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STUDIES ON METABOLIC DISORDERS IN DAIRY COWS

III. THE EFFECTS OF AFTER-CALVING UNDERFEEDING AND OF THYROPROTEIN DOSING ON THE LEVEL OF SERUM MAGNESIUM IN DAIRY COWS

By J. B. SWAN and N. D. JAMIESON, Animal Research Station, Department of Agriculture, Wallaceville

(Received for publication, 11 February 1956)

Summary

Clinical grass staggers has been produced at Wallaceville, a district where the disorder does not occur naturally, by underfeeding suitable cows with or without thyroprotein dosing.

Evidence has been obtained showing that the following conditions cause a lowering of the level of serum magnesium in milking cows:

1. Feeding thyroprotein alone or in conjunction with under-feeding.
2. A sudden change in quality and quantity of the ration.
3. A sudden increase in the quantity of the ration.
4. Calving, especially if accompanied by a reduction in grazing time.
5. Oestrus, if accompanied by marked homosexual activity and a disturbance of grazing behaviour.
6. Strong winds, where there is a reduction in grazing time or, if not, a marked reduction in ruminating time.

It is suggested that these conditions produce hypomagnesaemia by causing a negative energy balance, or digestive disturbances or both.

Clinical grass staggers may develop when one or more of these conditions act sufficiently strongly on susceptible cows, i.e., those that are in good condition; high producers; in the early weeks of lactation —particularly those in their second or third lactation season.

INTRODUCTION

As a result of the field observations reported in the previous paper (Swan and Jamieson 1956b), it was decided to test recently-calved cows under controlled conditions at Wallaceville for the effect of underfeeding on the serum magnesium level and the incidence of metabolic disorders.

Underfeeding has been produced in three forms in dairy cows by Eckles and Palmer (1916), namely: absolute underfeeding by deficient food intake; relative underfeeding, by sudden reduction from excessive

to adequate food intake; and "physiological" underfeeding, described as the condition arising in high-producing, good-conditioned cows in early lactation unable to ingest sufficient food to meet their high maintenance and production requirements. Of these, we have tested the effects of two, absolute and "physiological" underfeeding, simulating the latter by dosing iodinated casein.

EXPERIMENTAL

Methods

These have already been given for calcium, magnesium, and ketones (Swan and Jamieson 1956a).

Liveweights of the cows were determined directly after the evening milking, using an Avery platform scale. Milk weights were recorded at the bail using the apparatus of Whittleston and Verrall (1948). Milk fat was determined by the Gerber fat test, and serum alkaline phosphatase by the method of King and Armstrong (1934) using the sodium carbonate-bicarbonate buffer of Delory and King (1945).

Dry-matter intake was calculated from the faeces dry-matter excreted over 12-hour periods as determined by the chromium sesquioxide dosing technique, and the dry-matter digestibility coefficient from the faecal nitrogen content (Lancaster 1949; Lancaster *et al.* 1953).

The thyroprotein used in the dosing experiments was iodinated casein, prepared by Dr. I. L. Campbell at the Dairy Research Institute, Palmerston North, and later Protamone, a proprietary preparation.

Animal Management

Animals due to calve were, following normal Wallaceville practice, segregated from the herd a day or two beforehand and the cow and calf returned to the herd the next day. Methods of underfeeding varied slightly from one experiment to another but generally the restricted group was fed a daily allowance of pasture limited by an electric fence, or else the underfed group were limited to a field of grass grazed short by sheep, while the controls were rotated over a much larger area of similarly grazed pasture. In some of the earlier work, the cows were milked three times daily in an endeavour to impose a greater lactational strain. Blood samples were taken directly after the afternoon milking.

RESULTS

Table 1.—The effect on the serum magnesium levels of cows underfed while grazing short spring pasture in the third month of lactation.

Ten cows were grazed for 13 days continuously on a $1\frac{1}{2}$ acre paddock, while ten controls were rotated over 6 similar paddocks. The results (Table 1) show that it is the quantity rather than the stage of growth of the pasture that is the deciding factor in lowering serum magnesium. No clinical signs were observed in any of the underfed cows apart from three cases of mastitis. The cows showing the greatest fall in serum magnesium were those most recently calved, and those that continued to milk better than would be expected in view of the severe underfeeding imposed.

TABLE 1. EFFECT ON THE SERUM MAGNESIUM LEVEL OF UNDERFEEDING SHORT, SPRING PASTURE TO COWS IN THE THIRD MONTH OF LACTATION

	Serum Magnesium (mg.%)											Mean Serum Ca mg.%	Mean Milk Fat %	Mean Milk Wt. lb./day	Mean Days Calved	Mean Age
	Individual Cows										Mean					
Before underfeeding																
Rotated	3.0	2.6	2.6	2.8	2.5	2.4	2.5	2.5	2.7	2.6	2.6	10.4	5.18	24.0	100	6 yr.
Restricted	2.9	2.7	2.6	2.9	2.6	2.3	2.4	2.8	2.5	2.5	2.6	10.2	5.83	18.2	76	5 yr.
13th Day																
Rotated	2.5	2.5	2.7	2.6	2.4	2.3	2.4	2.4	2.7	2.7	2.5	11.0	4.87	22.6		
Restricted	2.3	2.3	1.8	2.1	2.3	2.2	1.5	2.3	2.0	1.7	2.0	10.9	6.18	15.9		
12 days Normal grazing																
Rotated	2.9	2.6	2.8	3.0	2.8	2.5	2.5	2.8	3.0	2.7	2.75	10.6	4.63	21.0		
Previously Restricted Group	2.8	2.7	2.4	2.8	2.8	2.4	2.4	3.0	2.7	2.8	2.7	10.7	4.82	26.0		

Controls—Rotational grazing over 9 acres—10 cows

Underfed—Continuous grazing on 1½ acres—10 cows

Table 2.—The effect on the serum magnesium levels of cows underfed within one week after calving.

Four cows were grazed on daily breaks of short pasture, previously grazed by sheep. They also received four pounds of hay daily and were milked three times daily to impose extra lactation stress.

D.8 and E.11 were both thin cows, but lost much condition, milked fairly well and developed a moderate ketosis and hypoglycaemia. D.8's rise in magnesium in the last week was accompanied by a decline in milk yield.

E.10 and E.1 were in fairly good condition but lost condition markedly. E.10 milked rather poorly, while E.1 maintained her lower level of production fairly well during underfeeding. Both had high fat tests during underfeeding—maximum of 7.1 and 7.4% fat—and both showed hypoglycaemia and ketosis, during the first week of underfeeding, which disappeared slowly. Both showed slight signs of muscular tremors and inco-ordination in the second week of underfeeding, on one day only. Body weight losses averaged 100 lb.

Table 3.—Thyroprotein dosing—The effect of thyroprotein dosing on the serum magnesium level of normal and underfed recently-calved cows.

Thyroprotein was dosed to 4 groups of underfed and normally fed cows to determine whether the loss in body weight, and increased milk and fat yield caused by this drug, would produce hypomagnesaemia in normally fed animals, and increase the hypomagnesaemia of underfed cows. The dose was administered as iodinated casein as a drench at evening milking each day at the rate of 30 g. per 1,000 lb. body weight.

The underfed cows in this experiment received only small daily breaks of spring pasture, 2 to 3 in. long. The controls grazed similar pasture *ad lib.* plus hay and turnips.

The results in Table 3 show that thyroprotein causes a marked reduction in serum magnesium in normally fed cows in early lactation, and that it causes a further fall in magnesium in cows on a restricted food intake.

All the dosed cows except D.7 gave increased milk and fat yields, accompanied by an increased pulse rate and rectal temperature, and a loss of body weight.

On the third day before the last dose, E.10 showed slight ataxia and muscular tremors.

Table 4.—Thyroprotein dosing combined with underfeeding and the production of clinical signs of tetany and paresis in cows in early lactation.

Eight cows were underfed, starting in the first week of lactation, and dosed thyroprotein at the rate of 30g. per 1,000 lb. body weight after evening milking each day. The cows were grazed in daily small breaks of short grass with no other feed. No control cows were included in this experiment owing to the small number of suitable animals available.

TABLE 2. EFFECT ON SERUM MAGNESIUM AND OTHER VALUES, OF UNDERFEEDING COMMENCING WITHIN ONE WEEK OF CALVING

	Prelim. Week		Two Weeks Underfeeding						Prelim. Week		Two Weeks Underfeeding					
	Serum Magnesium mg.%								Serum Calcium mg.%							
Underfed																
D8	2.0	2.3	1.7	1.3	1.2	1.4	2.0	1.9	9.8	10.2	8.8	9.6	9.6	9.5	9.8	9.7
E11	1.9	2.0	1.3	1.0	1.0	1.0	1.1	1.4	9.8	9.9	9.4	10.1	9.9	10.2	10.2	10.7
E10	2.3	2.4	1.8	2.1	1.7	1.8	1.6	1.1	9.5	9.3	8.5	9.7	8.9	9.2	9.4	9.3
E1	1.7	1.8	1.0	0.8	0.8	0.5	0.7	1.4	8.9	10.3	9.2	9.8	9.6	9.6	10.1	10.8
Mean	2.0	2.1	1.5	1.3	1.2	1.2	1.4	1.5								
Control																
D2	2.7	2.6	2.4	1.8	2.7	2.5	2.4	2.5	9.5	10.1	9.7	8.8	8.9	9.9	9.8	9.4
E3	2.9	2.6	2.2	2.3	2.6	2.5	2.6	2.6	9.8	9.2	9.5	9.9	9.9	10.1	9.6	9.9
D1	2.1	2.6	3.3	—	2.8	2.8	2.4	2.6	10.0	10.2	10.8	9.8	9.8	9.0	9.6	9.9
D13	2.5	2.7	—	2.3	2.3	2.7	2.5	—	9.8	9.3	—	9.5	9.7	10.1	10.0	—
Mean	2.6	2.6	2.6	2.1	2.6	2.7	2.6	2.6								
	Blood Glucose mg.%								Blood Ketones (Urine Rothera Tests) mg.%							
Underfed																
D8	42.5	44	46	38	38	38	55	47	(-ve)	(1+)	13	11	6	3.4	7	6
E11	—	49	31	46	48	51	—	55	(3+)	7	21	13	20	6	11	(1+)
E10	50	50	43	37	44	48	46	52	—	—	26	23	16	10	14	7
E1	—	58	28	47	39	52	—	55	—	—	19	5.5	21	7	—	—
Control																
D2	37	42	—	47	—	—	54	—	(-ve)	(-ve)	(±)	—	—	(4+)	8.5	—
E3	41	—	—	48	—	47	—	47	—	—	—	6.5	—	—	—	—
D1	47	—	—	—	51	42	38	51	12	—	—	—	(4+)	17	15	—
D13	47	—	—	—	54	—	41	—	—	—	—	(1+)	6.5	—	—	—
	Milk Weight lb./day								Milk Fat %							
Underfed																
D8	20.7	28.2	24.9	22.3	23.5	20.3	18.4	17.8	2.3	4.2	5.3	5.6	5.0	4.8	4.3	4.2
E11	34.9	35.1	35.2	33.6	37.8	34.4	28.6	33.2	4.2	5.1	4.6	3.7	3.7	4.1	4.1	4.2
E10	33.3	32.3	22.0	21.6	19.4	19.5	16.3	15.8	3.9	3.4	5.3	6.3	6.1	6.1	6.6	7.1
E1	23.5	22.4	17.0	20.2	15.8	17.5	14.4	16.1	4.9	3.4	5.4	7.4	5.9	5.6	5.3	5.7
Control																
D2	29.6	33.9	34.6	36.3	34.0	35.0	34.0	35.7	4.6	4.8	4.6	4.2	4.5	4.5	3.9	4.9
E3	28.5	29.5	29.8	25.1	29.8	31.4	29.9	30.9	3.4	3.8	4.1	4.3	4.2	4.3	4.2	4.1
D1	11.5	25.6	28.7	—	—	—	—	34.6	2.5	5.2	4.1	—	—	—	—	4.7
D13	28.9	—	—	24.5	34.4	—	31.4	—	3.3	—	—	3.5	3.6	—	3.5	—

TABLE 3. SERUM MAGNESIUM LEVELS THRICE WEEKLY IN INDIVIDUAL COWS

			Thyroprotein dosing on normal feed		Dosing started 7/9, ended 23/9								
Cow	Calved		Prelim. Week		During Dosing				After Dosing				
D.7	6/8		2.7	2.9	1.9	2.1	2.1	2.3	2.4	2.5	2.5	2.7	2.8
D.14	17/7		2.3	1.9	1.3	1.3	1.3	1.5	1.7	1.9	2.1	2.1	2.4
F.1	29/7		2.3	2.5	1.8	1.8	1.6	2.0	2.2	2.7	3.0	2.8	3.0
	Means		2.4	2.4	1.7	1.8	1.7	1.9	2.1	2.2	2.5	2.5	2.7
			Undosed Controls on normal feed										
D.2	23/7		2.7	2.5	2.7	2.7	2.6	3.0	2.5	2.7	2.7	3.0	2.9
D.13	3/8		2.5	2.9	2.7	2.6	2.6	3.0	2.6	2.5	2.0	2.3	2.1
E.7	9/8		2.5	2.5	2.5	2.4	2.2	2.4	2.5	2.4	2.4	2.4	2.5
	Means		2.6	2.6	2.6	2.6	2.5	2.8	2.5	2.5	2.4	2.6	2.5
			Thyroprotein dosing during underfeeding										
Cow	Calved	Under-fed from											
E.10	25/7	3/8	1.8	1.8	1.1	0.6	0.8	1.7	1.4	2.2	2.6	2.7	2.6
D.11	5/8	1/9	2.6	2.4	1.4	1.7	1.6	2.2	2.0	2.4	2.5	2.7	2.6
	Means		2.2	2.1	1.3	1.2	1.2	2.0	1.7	2.3	2.6	2.7	2.6
			Undosed controls during underfeeding										
D.8	25/7	3/8	2.0	1.5	1.6	1.9	1.4	1.9	1.7	2.0	2.2	2.5	2.1
D.15	10/8	7/9	2.0	2.3	1.9	1.8	1.8	2.3	2.2	2.3	2.3	2.4	2.3
	Means		2.0	1.9	1.8	1.9	1.6	2.1	2.0	2.2	2.3	2.5	2.2
Bleeding date			31/8	7/9	10/9	14/9	17/9	21/9	24/9	28/9	1/10	5/10	8/10

The symptoms shown by E.3, D.13, and D.8 at her second attack closely resembled those seen in natural cases of grass staggers. The attack of temporary paresis in D.1 and the mild symptoms in E.10 may be regarded as due to the treatment of this experiment. Natural cases of grass staggers have not been observed in the Wallaceville herd at any previous time or since.

All cows in this experiment were underfed for three or four days after calving before starting on the experimental underfeeding proper. This had been for years a standard practice at Wallaceville, the freshly-calved cows being kept in a bare paddock near the milking-shed until they were taken into the herd. This practice was doubtless responsible for the hypomagnesaemia noted in D.1, D.2, E.10, E.11, and F. 4 before experimental underfeeding began.

Most of the cows milked fairly well during underfeeding, at least until clinical symptoms of staggers occurred. Body-weight losses were of the order of 100 to 150 lb. up to the occurrence of symptoms in E.3, D.13, and E.10, and 200 lb. in D.1. F.4 lost 100 lb., E.11 and D.2 lost 50 lb. over the same period. About half of this weight loss was regained a week after normal feeding was resumed, indicating that this much at least of the loss was due to decrease in gut-content.

There may be some significance in the fact that E.3 and D.13 were the fattest cows used in this experiment, while the only thin cow to show symptoms was D.8. D.1 and E.10 were in fairly good condition, D.2, E.11, and F.4 fairly thin.

The important factor in the development of the grass staggers cases, E.3, D.13, and D.8 (2nd attack), would appear to be hypocalcaemia, and whether hypomagnesaemia plays more than a contributory part in producing the symptoms is open to doubt. It is noteworthy that E.3 and D.13 both responded immediately to glucose and calcium injection on their first attacks. It was intended to give magnesium also at this time, but this did not appear necessary.

The influence of thyroprotein dosing in the production of these cases may have been due solely to the action of this drug in stimulating milk production at the expense of the body tissues, or it may have had some action on the parathyroids, thus producing the hypocalcaemia. Dogs which had been dosed "Protamone" for several weeks, and were then subjected to thyroidectomy and partial parathyroidectomy, developed tetany which did not respond to calcium or parathyroid injection, suggesting that thyroprotein may antagonize the parathyroid. Similar operations on untreated dogs did not cause tetany (Borgman and Reinecke 1950).

As a result of the previous findings, it was decided to test the effect of underfeeding alone on cows in good bodily condition in the first week or two after calving, and to estimate the degree of underfeeding applied, by measurement of food intake over the period.

Tables 5—Controlled underfeeding experiments:

Eight cows in the first to second week of lactation were changed suddenly from a ration of spring pasture plus hay and kale to limited pasture alone at three different levels of intake for periods of from ten to thirteen days. The first group of two cows, in fair condition, were

TABLE 4. DAILY BLOOD ANALYSES ON INDIVIDUAL COWS

Cow E.3. Calved 22/7, underfed 9-26/8, thyroprotein 12-24/8

Date: August	2	5	9	12	15			16	18	19	23	30
					10 a.m.	2 p.m.	4 p.m.					
Serum Ca	9.6	9.9	9.3	9.9	7.0	6.6	7.0*	8.7	5.1*	7.8	9.6	10.4
Serum Mg	2.4	2.1	2.3	1.1	0.4	0.3	0.3	0.4	0.3	0.9	0.7	1.9
Blood Sugar					63	128	138		105			
Blood Ketones				12	16	7	6		3			
					Tetany & paresis				do.			

*Treated 15/8 4 p.m.: Glucose and calcium gluconate, recovered. 18/8 calcium gluconate and magnesium sulphate. Recovered.

Cow D.13 Calved 26/7, underfed 9-26/8, thyroprotein 12-24/8

Dates as above												
Ca	10.3	10.2	10.5	10.4		5.6	6.4*	8.1*	6.6	6.7	9.7	10.3
Mg	2.6	2.6	2.5	0.7		0.1	0.15	0.4	0.9	0.7	0.9	1.4
Sugar						168	273		45			
Ketones				13		5	5		11	20	10	
						Tetany & paresis						

*Treated 15/8 4 p.m.: Glucose and calcium gluconate, recovered. 16/8 calcium gluconate and magnesium sulphate. Recovered.

Cow D.8. Calved 2/9, underfed 2-6/9, thyroprotein 5/9 only

Date:	30/8	3/9	6/9			7/9	9/9	13/9
			5 a.m.	11 a.m.	4 p.m.			
Ca	11.1	5.1*	3.8*	8.4*	7.9*	8.0	9.6	9.8
Mg	2.0	2.3	1.4	2.9	2.8	1.1	1.4	1.9
Sugar			127	69	95	63		
Ketones		2	2	1	6			
		Ataxia	Tetany & paresis					

*Treated 3/9 2 p.m. Calcium gluconate. Recovered promptly.
6/9 5 a.m. Calcium gluconate, magnesium sulphate and glucose. Rose, but fell again and stayed down.
11 a.m. Udder inflated. No response.
4 p.m. Calcium gluconate. No response. Eating.
7/9 Eating well, but weak and still down.
9/9 Able to walk when lifted. Eating well.

Cow E.10. Calved 5/8, underfed 12-26/8, thyroprotein 12-24/8.

Date: August	9	12	16	19
Ca	9.6	10.0	10.4	9.5
Mg	1.7	2.0	1.5	0.9

19/8: Mild symptoms ataxia and muscular tremors for 15 minutes.
21/8: Injured during service—very lame, turned out.

Cow D.1. Calved 21/8 underfed plus thyroprotein 26/8-6/9.

Date: August	9	23	26	30	Sept. 2	6	9	13
Ca	10.0	9.8	7.5	10.9	9.2	9.3	9.4	9.7
Mg	2.3	2.0	1.0	0.8	1.0	1.7	1.5	2.2

7/9: Temporary paresis 5-7 a.m. Recovered spontaneously.

Cow E.11. Calved twins 25/7, underfed 12/8-6/9, thyroprotein 19-24/8.

Date: August	2	5	9	12	16	19	23	26	30
Ca	9.3	9.5	9.4	9.9	10.2	10.0	9.8	10.1	10.0
Mg	1.8	1.8	1.4	1.5	0.7	1.1	0.8	0.7	1.5
Ketones				5					

Very thin, lost condition after calving, retained placenta.

Cow D.2. Calved 5/8, underfed 12/8-6/9, thyroprotein 12-24/8.

Date: August	9	12	16	19	23	26	30	Sept. 6	9	13
Ca	10.9	9.9	10.1	9.9	9.6	9.7	9.5	9.4	9.7	9.7
Mg	1.6	2.3	2.2	2.0	2.1	2.1	2.4	2.7	2.3	—

Cow F.4. Calved 3/8, underfed 12/8-6/9, thyroprotein 12-24/8.

Dates as above										
Ca	10.8	10.0	10.5	9.5	10.1	10.1	10.0	9.6	9.9	9.7
Mg	1.6	2.4	1.9	1.5	1.5	1.5	2.3	2.4	2.5	2.5

TABLE 5A. COWS IN FAIR CONDITION. MILD UNDERFEEDING FOR 10 DAYS

Days 1st day 19.7.50	J.45 Friesian, 4 yr. Calved 12.7.50 — Underfed 6-15th day inclusive							K.10 Jersey, 3 yr. Calved 9.7.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb	Serum Ca mg.%	Serum Mg mg%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	900	10.5	3.1	7.1	15.5	6.2	—	632	10.6	2.7	6.4	17.3	4.8	—
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—
3	—	10.6	3.1	7.4	17.8	6.5	23.8	—	11.3	3.2	7.1	19.0	5.3	22.9
4	—	—	—	—	—	—	20.2	—	—	—	—	—	—	19.5
5	—	10.1	2.5	6.0	10.2	5.0	24.6		10.7	3.1	7.3	17.4	4.1	17.8
6	824	10.0	2.3	5.7	13.2	7.0	13.1	644	10.7	3.1	6.9	19.0	5.3	18.3
7	—	9.7	2.7	6.2	(10.6)*	6.4	9.6	—	10.3	3.2	6.2	20.6	5.2	18.0
8	790	9.9	2.6	5.7	(10.3)	6.8	8.9	640	10.7	3.0	6.2	18.5	4.2	17.9
9	—	9.9	2.5	5.2	(10.2)	4.6	6.3	—	10.7	2.9	6.0	19.5	4.5	17.8
10	—	9.3	2.2	5.0	(8.3)	6.7	7.0	644	10.4	3.0	6.3	19.4	5.6	16.5
11	—	9.6	2.1	4.7	(9.3)	5.8	7.9	—	10.4	2.9	6.6	19.6	4.6	19.1
12	—	9.9	2.3	4.8	(11.0)	4.9	10.4	—	10.2	2.9	6.1	19.8	4.9	19.8
13	780	10.3	2.3	5.2	(9.8)	5.2	8.1	628	10.3	2.9	5.9	18.5	4.1	21.6
14	—	10.3	2.4	5.2	(11.7)	4.9	8.0	-	10.9	3.1	6.7	19.4	8.0	16.9
15	760	10.0	2.4	4.9	(10.8)	5.4	8.7	620	10.0	3.1	6.4	18.0	5.4	17.2
16	—	10.3	2.5	5.7	(10.3)	5.1	8.2	—	10.7	2.8	7.0	17.5	4.6	14.7
17	788	10.4	2.2	4.5	(11.2)	4.2	18.4	628	10.2	2.6	6.5	18.3	4.2	21.8
18	—	—	—	—	(12.2)	5.0	13.2	—	—	—	—	18.3	4.9	23.3
19	—	—	—	—	(12.7)*	4.5	16.9	—	—	—	—	17.3	4.8	20.8
20	794	10.0	2.7	5.6	13.0	5.9	18.2	614	10.1	2.7	6.1	17.2	4.0	16.9

Days 1st day 19.7.50	D.2 Jersey, 9 yr. Calved 18.7.50 — Underfed 6-15th day inclusive							F.4 Jersey, 7 yr. Calved 5.7.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb.	Serum Ca mg.%	Serum Mg mg%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	—	7.7	2.6	8.6	—	—	—	-	10.0	2.5	9.5	30.9	5.3	—
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—
3	—	10.5	2.2	9.4	30.4	5.1	22.9	—	10.5	2.7	9.1	30.3	7.1	26.9
4	—	—	—	—	—	—	29.9	—	—	—	—	—	—	29.3
5	—	9.8	2.4	8.3	32.4	5.3	25.6	—	9.9	2.8	10.0	31.6	4.6	24.2
6	808	9.3	2.5	7.0	32.8	6.1	28.4	838	9.5	2.6	10.1	30.3	4.9	25.3
7	—	9.2	2.5	7.7	29.9	6.2	15.0	—	9.5	2.9	8.6	30.3	5.1	22.4
8	768	9.7	2.4	7.8	25.7	6.6	18.5	832	10.3	2.8	9.2	30.9	5.0	26.4
9	—	9.4	2.1	7.4	27.0	6.3	11.0	—	10.0	2.8	8.4	33.2	5.1	21.0
10	790	9.2	2.0	6.5	28.4	6.4	14.2	832	9.7	2.5	9.2	31.3	5.1	23.9
11	—	9.3	2.0	7.6	26.7	5.6	18.3	—	9.7	2.7	10.0	32.3	4.7	27.2
12	—	9.6	1.9	8.2	29.1	6.0	16.7	—	10.0	2.8	9.2	32.4	4.5	24.2
13	776	9.9	1.9	9.1	29.3	6.0	19.1	836	9.9	2.7	8.8	34.2	4.1	30.0
14	-	9.8	1.8	10.3	25.4	5.4	14.9		10.7	3.0	8.1	29.7	3.9	26.7
15	760	9.3	2.2	9.6	25.9	6.0	14.6	834	9.9	2.9	8.4	34.1	4.8	31.3
16	—	10.1	2.2	9.8	27.2	5.3	14.5	—	9.8	2.9	8.1	30.9	3.8	33.6
17	764	10.2	1.9	13.1	31.9	5.8	49.4	828	10.0	2.8	7.9	31.2	4.0	25.7
18	—	—	—	—	32.0	5.1	33.4	—	—	—	—	26.8	4.5	26.6
19	—	—	—	—	31.2	4.4	24.3	-	—	—	—	33.3	6.1	25.2
20	768	9.6	2.5	11.4	31.9	4.7	25.4	822	9.8	2.4	7.5	28.5	4.5	22.3

* Mastitis (Milk from three quarters only)

TABLE 5B. COWS FAIRLY GOOD CONDITION. MODERATE UNDERFEEDING FOR 10 DAYS

Days 1st day 8.8.50	J. 11 Friesian 4 yr. Calved 6.8.50 — Underfed 6-15th day inclusive							J.8 Friesian, 4 yr. Calved 5.8.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	1141	10.0	2.3	7.7	38.1	6.1	—	972	9.4	2.2	14.7	20.7	—	—
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—
3	1158	11.0	1.8	8.9	38.7	5.9	27.2	982	10.0	2.1	15.1	24.6	4.2	29.3
4	1160	—	—	—	—	—	29.1	989	—	—	—	35.5	—	34.1
5	1189	9.9	2.6	8.8	42.6	5.5	29.4	999	10.0	2.8	13.3	—	—	31.4
6	1122	—	—	—	35.8	5.9	30.0	978	—	—	—	32.4	—	33.0
7	1075	10.0	2.1	6.4	32.4	6.0	17.9	1008	10.3	2.3	13.1	37.7	3.5	30.1
8	1026	9.6	2.2	7.7	30.8	6.8	16.9	981	9.8	2.3	12.0	33.7	3.6	30.4
9	1021	9.8	2.2	6.0	31.0	6.4	13.3	995	9.8	2.5	14.8	39.0	3.4	31.1
10	996	10.1	2.2	5.5	25.7	6.9	21.3	966	10.1	2.4	13.4	33.5	3.7	29.0
11	997	9.9	1.9	5.3	29.1	7.7	11.3	977	10.1	2.3	16.2	40.8	3.8	35.5
12	975	10.2	1.6	6.0	23.5	7.5	15.7	960	9.7	2.3	13.7	35.2	4.0	34.0
13	963	10.8	1.6	5.5	24.6	6.8	14.1	956	10.4	2.3	12.7	40.8	3.5	30.5
14	957	10.8	1.5	5.5	28.0	6.8	16.6	996	10.3	2.3	12.3	41.6	3.4	36.8
15	939	10.1	1.5	4.8	23.6	6.3	13.7	955	10.1	2.5	14.0	39.8	3.8	41.2
16	987	10.9	1.5	6.5	26.8	6.1	20.6	971	10.6	2.5	13.0	36.2	4.3	36.2
17	972	10.8	1.1	6.9	35.8	5.3	21.4	991	10.0	2.0	13.4	40.0	4.0	36.3
18	998	10.6	1.2	8.0	37.2	4.8	16.9	980	10.5	2.0	11.5	39.0	3.5	39.6
19	1039	11.3	1.6	9.4	37.0	4.4	28.4	982	10.3	2.2	12.6	41.4	3.4	39.9
20	1028	10.8	1.9	8.7	38.2	4.2	29.0	982	10.0	2.0	11.7	40.8	3.2	41.6

Days 1st day 8.8.50	K.9 Jersey, 3 yr. Calved 24.7.50 — Underfed 6-15th day inclusive							J.48 Friesian, 4 yr. Calved 5.8.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	684	10.7	2.4	5.0	21.0	5.8	—	—	10.4	2.4	6.0	10.4	3.8	—
2	—	—	—	—	—	—	—	882	—	—	—	—	—	—
3	684	11.5	2.6	5.5	21.6	6.7	17.1	867	11.0	2.2	8.3	11.9	5.7	15.3
4	705	—	—	—	—	—	17.7	882	—	—	—	—	—	19.2
5	786	10.6	3.6	6.5	19.5	7.4	23.3	885	9.9	3.3	7.2	17.4	8.7	20.8
6	668	—	—	—	16.1	5.6	19.9	873	—	—	—	11.5	4.5	12.9
7	628	10.8	2.3	6.6	22.4	6.4	12.1	892	10.3	2.8	6.9	19.4	7.3	21.6
8	624	9.9	2.3	5.6	15.2	7.8	6.9	871	9.6	2.7	7.1	15.3	6.2	18.4
9	642	10.2	2.2	5.5	14.1	5.8	7.1	869	10.2	2.9	7.2	18.5	6.3	24.6
10	606	10.3	2.0	5.3	13.3	5.9	9.8	863	10.3	2.8	6.3	19.7	5.9	17.2
11	610	10.0	1.9	5.8	12.0	4.3	15.2	860	9.8	2.7	6.1	18.0	5.8	16.2
12	606	9.9	1.8	5.5	15.9	8.8	15.8	845	9.8	2.6	6.9	19.1	5.7	18.3
13	591	10.7	1.9	5.6	13.6	5.7	14.0	862	10.3	2.5	7.2	19.2	5.0	23.5
14	607	10.7	1.7	4.5	10.8	3.4	15.0	852	10.3	2.6	6.3	17.4	3.6	18.7
15	596	9.4	1.9	4.6	13.0	5.5	14.4	841	10.1	2.6	6.9	16.6	4.2	21.8
16	635	11.1	2.0	5.3	14.4	9.2	11.1	844	10.5	2.4	6.7	16.6	4.3	18.5
17	630	10.7	1.5	5.0	15.2	5.3	12.1	858	11.0	2.4	7.4	19.8	5.8	15.2
18	646	11.4	1.5	6.2	15.6	4.8	15.4	854	10.4	2.4	7.4	14.2	4.2	20.0
19	669	11.1	1.9	6.3	18.0	4.5	19.1	866	10.4	2.5	7.4	14.6	4.0	24.0
20	675	10.5	2.3	6.0	17.2	4.6	20.7	850	10.4	2.4	6.6	18.2	5.1	21.1

TABLE 5C. COWS IN GOOD CONDITION. SEVERE UNDERFEEDING FOR 13 DAYS

Days 1st day 8.9.50	E.3. Jersey, 8 yr. Calved 6.9.50 — Underfed 6-18th day inclusive							E.11. Jersey, 8 yr. Calved 18.8.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	1045	9.8	2.4	6.0	20.3	3.8	—	836	10.7	2.1	27.0	28.2	5.0	—
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—
3	1068	9.7	2.0	6.0	24.2	4.4	23.1	851	10.3	2.1	29.0	27.9	5.2	38.3
4	1084				—	—	25.6	863	—	—	—	—	—	23.8
5	1040	8.7	2.4	6.6	27.8	4.6	25.0	848	10.0	2.0	30.0	29.6	4.7	33.7
6	1000	6.1	2.4	4.7	22.2	5.2	19.5	837	10.3	2.0	30.0	25.6	5.1	18.9
7	972	8.3	2.4	4.7	19.8	—	17.3	843	10.4	2.0	28.5	28.2	—	23.4
8	950	9.3	1.7	4.5	17.8	5.1	11.2	860	10.9	2.2	25.0	29.6	4.5	25.6
9	931	8.6	1.5	3.7	18.4	6.7	9.6	871	10.7	2.1	27.0	29.2	4.5	32.4
10	912	8.7	1.2	4.3	15.4	7.5	6.4	849	10.1	2.1	28.0	27.6	4.6	26.2
11	899	7.7	1.0	4.0	15.6	6.9	5.3	877	10.1	2.4	26.0	28.4	4.4	21.2
12	904	7.5	0.9	3.9	14.6	7.8	4.5	866	10.2	2.2	24.0	30.8	4.3	29.8
13	886	7.9	0.9	3.6	15.6	6.6	5.6	843	9.6	2.1	25.0	30.0	—	35.8
14	848	7.4	0.9	3.7	11.4	6.4	4.7	832	10.3	2.0	24.0	29.4	4.8	22.0
15	859	7.2	0.8	4.3	12.8	8.2	5.3	823	10.8	1.9	25.0	29.0	4.4	17.5
16	855	7.4	0.7	3.6	14.2	6.8	5.7	827	10.6	2.0	24.0	33.0	4.5	22.8
17	845	7.4	0.9	4.6	14.4	6.0	5.1	847	10.3	2.3	24.5	30.8	4.5	30.8
18	844	7.3	1.0	4.9	14.4	6.5	5.7	827	10.5	1.8	27.0	29.4	4.4	20.2
19	875	8.9	1.0	5.1	15.6	4.5	8.8	840	11.0	2.0	26.0	29.0	4.3	33.8
20	883	10.2	0.9	5.2	18.8	5.8	12.6	847	10.5	1.9	27.0	32.2	4.1	26.7
21	896	9.9	1.0	6.2	20.2	5.1	14.4	840	10.2	1.9	25.5	33.6	4.1	29.5
22	915	10.3	1.6	6.9	23.2	4.8	20.3	847	10.3	2.1	26.0	30.4	3.8	22.0
23	925	9.3	1.9	6.5	21.2	3.9	25.2	854	9.5	1.9	24.5	31.8	4.4	28.6

Days 1st day 8.9.50	D.13. Jersey, 9 yr. Calved 26.8.50 — Underfed 6-18th days inclusive							J.6. Friesian, 4 yr. Calved 12.8.50 — Control						
	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day	Live weight lb.	Serum Ca mg.%	Serum Mg mg.%	Serum Alkal. Phosphatase K.A. units	Milk weight lb.	Milk Fat %	D.M. Intake lb./day
1	1008	10.8	2.1	7.2	37.5	5.0	—	961	10.2	2.4	6.6	36.2	3.6	—
2	—	—	—	—	—	—	—	—	—	—	—	—	—	—
3	971	10.1	2.3	7.2	39.2	5.1	25.0	912	10.4	2.3	8.0	38.7	3.8	—
4	984	—	—	—	—	—	25.5	938	—	—	—	—	—	29.0
5	946	10.2	2.0	7.0	39.4	5.4	25.1	912	9.9	2.2	8.0	39.0	3.4	27.7
6	908	9.6	1.9	6.6	21.6	5.8	27.2	913	9.6	2.1	7.0	32.2	4.1	23.4
7	904	9.9	2.0	7.5	33.9	—	15.8	931	9.9	2.1	5.9	37.8	—	28.7
8	881	9.9	1.6	6.0	25.0	5.2	10.5	927	11.1	2.2	6.8	38.8	3.3	29.6
9	866	9.9	1.6	6.6	23.4	6.2	14.6	941	10.1	2.3	6.5	38.4	3.1	26.5
10	848	9.9	1.7	7.1	19.6	6.6	10.7	903	10.4	2.4	7.4	39.6	3.9	28.7
11	820	9.7	1.7	5.8	20.6	6.6	12.2	954	9.8	2.4	7.3	35.8	3.2	30.6
12	825	9.4	1.7	5.8	21.4	6.2	9.6	944	10.3	2.5	7.3	41.8	3.2	28.0
13	810	9.8	1.9	6.8	20.0	4.9	9.7	924	10.3	2.4	7.0	38.6	3.6	34.1
14	805	9.6	1.8	7.9	18.8	6.7	12.5	928	9.8	2.3	6.6	35.0	3.0	27.3
15	799	9.7	1.7	7.2	18.6	5.9	8.8	907	10.3	2.1	6.6	38.4	3.3	26.0
16	806	10.2	1.7	8.0	18.0	4.7	8.5	912	10.4	2.3	(7.4)	40.0	3.3	24.3
17	812	10.3	1.9	7.7	21.4	(5.1)	8.9	918	10.2	2.5	6.6	42.4	3.5	31.1
18	787	9.3	1.7	7.8	20.0	4.8	13.7	910	10.2	2.3	7.0	37.6	3.0	27.4
19	847	10.6	2.0	8.5	20.0	4.5	11.9	937	10.1	2.3	7.0	40.0	3.0	30.1
20	856	10.4	1.4	7.8	27.2	4.6	13.6	916	10.0	2.3	6.7	42.8	2.9	29.0
21	855	10.9	1.7	8.8	28.0	4.3	20.8	909	10.4	2.4	7.6	43.0	3.0	30.1
22	873	10.9	1.8	10.1	32.0	4.9	30.4	926	10.7	2.3	8.3	41.8	3.2	32.7
23	879	10.6	2.2	8.7	32.2	4.4	30.4	898	10.6	2.3	7.4	39.4	3.6	25.0

"mildly" underfed in mid July for ten days (Table 5A). The second pair, in fairly good condition, were "moderately" underfed in mid August for ten days (Table 5B). The third group consisted of two cows in good condition plus two thin cows (whose results have not been given in detail), and was "severely" underfed in mid September for thirteen days (Table 5C). Two normally fed cows were run as controls in each experiment.

The cows were weighed and sampled daily, and their grazing behaviour observed on alternate days after underfeeding began.

Little or no loss of appetite occurred, as it appeared to have done a day or two before grass staggers symptoms occurred in the cows in the previous year's experiment. Rumination time was dramatically reduced, by as much as 90%, presumably due to lack of both bulk and fibre in the rumen. All cows developed diarrhoea after about three days of underfeeding, and strands of partly digested grass-leaf midrib were passed in the faeces.

The mildly underfed cows showed a drop of approximately 50% in food intake, and a live-weight loss of about 10%; those moderately underfed, a drop in food intake of about 60%, and a 25% live-weight loss. Food intake in the severely underfed cows fell by about 75% while the live weight decreased by only 20%. It would appear that the decrease in food intake is of greater import to the blood magnesium level than is the fall in live weight. Under the conditions of this experiment, the lowering of blood-magnesium levels to values below 1.5 mg.% occurred only if the restricted food intake fell below 75% of that obtained by unrestricted grazing.

Body weight decreased in proportion to the severity of underfeeding, much of the decline being due to gut-content decrease. Weights returned to the pre-underfeeding level in most cases within three weeks after returning to normal feed.

The "mildly" underfed cows showed only slight falls in serum magnesium, from 2.5 and 2.4 down to 1.8 and 2.1 mg.%. More marked falls, from 3.6 and 2.6 down to 1.5 and 1.1 mg.% occurred in the "moderately" underfed cows.

These levels are below the "normal" lower limits. The "severely" underfed cows suffered the most lowering of serum magnesium; the two fat cows fell from 2.4 and 2.0 to 0.7 and 1.4 mg.%, while the two thin ones, D.2 and D.8, fell from 2.5 and 2.0 down to 1.9 and 0.5 mg.%.

Slight hypocalcaemia also occurred in one fat and one thin cow in this experiment, levels ranging from 6.1 to 7.2 mg.% in the fat cow throughout the underfeeding period, and between 8.7 and 8.8 in the thin cow about the middle of the period, when hypomagnesaemia was most marked.

An unexpected finding was that serum magnesium levels in all underfed cows showed a tendency to a slight secondary fall on the second day after return to normal rations. In four cows, their lowest levels during the course of the experiment were recorded on that day.

The serum calcium and magnesium levels in the control cows remained within the normal range, with the exception of one cow which had a slight hypomagnesaemia (1.8 to 1.9 mg.%) on three occasions.

Milk and butterfat production were reduced in proportion to the level of underfeeding, and recovery to the expected normal production took place within the month following return to normal feed.

No clinical signs due to the hypocalcaemia or hypomagnesaemia appeared in any cow in this experiment, the only abnormality being a slight depression in the first few days, and in a few cows, especially one thin cow, towards the end of the underfeeding period.

Halse (1948) has reported a positive correlation between serum magnesium and serum alkaline phosphatase levels. Deficient dietary magnesium was not the cause of the hypomagnesaemia, and hypomagnesaemia was not the cause of the fall in serum phosphatase. Halse suggests the influence of some other factor disturbing the normal energy supply, which may be capable of affecting serum magnesium and phosphatase simultaneously.

The results in Table 5 show a fall in serum alkaline phosphatase concurrently with the fall in serum magnesium. Animal D.2 in Table 5 is anomalous in that the fall in serum phosphatase precedes that of the magnesium, and at the lowest magnesium level the alkaline phosphatase is rising.

On re-feeding, the serum phosphatase values returned to normal more rapidly than did the magnesium values.

It was thought that the failure to produce clinical signs of grass staggers in the cows of this last experiment without the use of thyroprotein was due to failure to underfeed sufficiently severely.

Table 6—A group of four cows severely underfed for two weeks during September, commencing in the first week of lactation.

A fairly successful attempt was made to have all the cows in good condition at calving, although feed was rather short for the last two weeks of the dry period. No hay or crops were fed during the dry period, as ample pasture was available at the start. On entering the herd two or three days after calving, hay and kale and a very little grass were fed for two or three days before the underfeeding experiment started. The control cows remained on this ration, there being a severe shortage of pasture in spring 1951, while the underfed cows were grazed on sparse pasture 1 to 2 in. long on daily breaks, allowing 88 sq. yd. per cow per day. This was about half the allowance given to the first group of cows underfed the previous year. The paddock was badly pugged during the winter and carried mostly ryegrass and a little clover.

One of the four underfed cows developed symptoms of milk fever and went down on the 5th day of underfeeding. She was swaying in the hindquarters and frothing up saliva by chewing movements at 9 a.m. By 2 p.m. she was prostrate but still able to lift her head. She was then treated by udder inflation. At 5 p.m. she was sitting fairly normally and, when approached, got up and walked away rather weakly and unsteadily. She was put back in the herd on hay, turnips, and short grass, made an uneventful recovery, and gave 46 lb. milk daily. Her weight on 15 September was 1016 lb. She was the thinnest of the underfed cows, being a high producer in the previous season (350 lb. fat in 300 days).

TABLE 6. 1951 Underfeeding Experiment

Days After Calving	Means Four Control Cows				Underfed Cows D.13				J.6			
	Live Wght. lb.	Milk Wght. lb.	Serum Ca mg.%	Serum Mg mg.%	Live Wght. lb.	Milk Wght. lb.	Serum Ca mg.%	Serum Mg mg.%	Live Wght. lb.	Milk Wght. lb.	Serum Ca mg.%	Serum Mg mg.%
0												
1												
2	900		8.4	1.8	957		10.5	1.7	1002		9.2	2.3
3	899		9.1	1.6	965	14.8	11.5	1.5	1033	37.4	9.9	2.0
4	899	20.8	10.4	1.5	912	18.6	9.1	1.3	952	29.8	9.2	1.9
5	900	19.9	10.5	1.6	899	15.8	7.9	1.2	920	27.4	9.1	1.9
6	902	22.6	10.2	1.9	887	16.4	8.1	1.2	925	24.6	8.4	1.5
7	897	24.7	9.8	2.2	880	15.6	11.7	0.9	947	27.0	10.0	1.0
8	907	26.0	9.7	2.2	866	17.4	10.1	1.0	856	33.4	8.9	1.0
9	904	25.6	9.9	2.4	851	15.6	9.4	1.0	831	25.4	8.2	1.1
10	899	26.9	10.1	2.4	844	13.6	9.2	1.1	813	20.2	7.6	1.0
11	906	26.2	9.7	2.3	853	16.0	10.0	1.2	813	18.4	7.3	1.1
12	907	23.5	9.9	2.4	827	16.4	10.8	1.3	817	21.2	8.7	1.4
13	901	23.4	9.7	2.1	820	17.8	10.7	1.4	793	21.2	9.2	1.5
14	897	21.1	9.7	2.3	822	18.0	9.4	1.4	812	18.8	8.5	1.7
15	906	21.3	9.2	2.2	815	15.4	9.1	1.2	812	21.2	8.2	1.7
16	892	22.7	9.8	2.2	815	7.8	9.3	1.8	798	19.0	9.5	1.7
17	895	23.4	9.6	2.2	786	7.8	9.8	1.5	765	16.6	9.0	1.6
18	884	24.5	9.5	2.2	788	6.8	9.7	1.4	756	12.8	8.9	1.4
19	880	23.7	9.5	2.1	793	9.0	9.4	1.4	764	14.4	8.8	1.3
20	876	23.6	10.1	2.1	856	9.8	10.2	1.5	835	15.6	9.7	1.4
21	885	26.7	9.9	1.9	837	14.0	9.2	1.6	820	19.2	9.9	1.2
22	875	26.8	9.7	2.1	866	14.2	9.7	1.8	837	22.2	9.8	1.4
23	893	27.7	9.6	2.2								
24	899	—	9.7	1.9	856	11.0	9.4	2.2	873	26.6	9.9	1.6
25	887	27.6	9.5	2.4	824		10.2	2.1	832		9.5	1.9
26					869	10.8	8.8	2.2	876	27.8	9.1	2.0
27	892	30.7	8.6	2.4								
28												
29												
30												
31	887	27.7	10.0	2.2	907	19.4	9.8	2.6	901	36.4	9.6	2.4
32												
33												
34												

Four Control Cows

	E.3	F.4
Age	9 yrs.	8 yrs.
Calved	27 Aug.	26 Aug.
Breed	J	J
Live wt.	970	830

	K.2	D.2
Age	4 yrs.	10 yrs.
Calved	25 Aug.	29 Aug.
Breed	F	J
Live wt.	996	796

D.13 Jersey

10 years
Calved 26 August
Underfed 4-19th days inclusive.

Mild Ketosis 15-18th days Serum Ketones 6 mg.% on day 16. Urine Rothera 5+ Slight ataxia on 17th day.

J.6 Friesian

5 years
Calved 26 August
Underfed 4-19th days post-calving inclusive.

Anorexia on 16th day. Moderate Ketosis 15-22nd days. Serum Ketones 54 mg.% on day 17. Urine Rothera 20+ Spastic tremors on 17th day.

Days After Calving	J.8 Live Wght. lb.	J.8 Milk Wght. lb.	J.8 Serum Ca mg.%	J.8 Serum Mg mg.%	J.10 Live Wght. lb.	J.10 Milk Wght. lb.	J.10 Serum Ca mg.%	J.10 Serum Mg mg.%	J.12 Live Wght. lb.	J.12 Milk Wght. lb.	J.12 Serum Ca mg.%	J.12 Serum Mg mg.%
0												
1												
2	993		10.2	2.1								
3	1013	38.0	10.9	1.9								
4	958	28.8	9.5	1.8					1054		9.6	1.7
5	926	32.2	11.0	1.5	1131		9.3	1.8	1104	32.8	9.6	1.7
6	838	34.4	8.4	1.3	1117	34.8	10.2	1.7	1096	27.0	10.2	1.9
7	830	24.2	7.2	1.4	1035	31.4	8.8	1.9	1089	34.0	10.0	2.2
8	—	17.8	5.8 5.6 7.5	1.4 1.4 1.8	971	17.4	7.7	1.9	1081	38.0	9.9	2.0
9	877	—	7.4	1.7	991	23.0	8.0	1.7	1077	42.2	10.3	2.5
10	857	(11.8)	9.1	1.5	1017	27.0	10.4	1.3	1076	39.6	10.6	2.3
11	877	22.0	9.8	1.6	914	32.4	8.4	1.2	1111	39.0	10.7	2.5
12	936	27.8	9.4	1.9	888	16.0	8.0	1.3	1099	41.8	10.4	2.2
13	932	31.0	8.2	1.8	878	25.8	8.4	1.4	989	32.6	9.2	1.7
14	—	33.4	—	—	883	18.4	8.2	1.7	946	24.2	9.1	1.6
15	919	31.6	10.5	2.3	891	20.0	9.1	1.8	932	20.6	8.4	1.4
16	—	35.2	—	—	885	21.6	9.3	2.0	922	21.0	9.3	1.5
17	—	35.4	—	—	888	20.2	9.2	2.0	917	29.8	9.1	1.5
18	1016	39.0	10.0	2.5	908	21.2	10.0	2.2	902	23.8	10.1	1.7
19	—	39.4	—	—	888	21.2	9.6	2.2	881	20.4	9.2	1.6
20	930	38.0	—	—	868	19.2	9.1	2.8	857	18.4	8.4	1.3
21	—	—	—	—	852	15.4	7.0	2.2	855	16.2	9.3	1.3
22	940	45.0	10.7	2.3	860	14.0	8.6	2.3	936	22.4	10.5	1.0
23	949	—	8.2	2.4	943	15.8	9.3	1.6	924	27.8	10.7	1.0
24	969	45.2	9.3	2.3	954	19.0	8.2	1.3	950	29.8	9.9	1.1
25					940	22.0	8.4	1.5				
26									995	35.6	10.0	1.7
27					990	26.0	8.8	2.2	964		10.3	1.6
28					983		9.9	2.2	1024	35.4	9.8	2.0
29	—	52.2	—	—	1023	31.6	9.2	2.2				
30												
31												
32												
33									1006	31.8	9.9	2.1
34					1028	43.8	9.8	2.4				

J.8 Friesian

5 years
Calved 28 August
Underfed 4-8th days post calving.

Milk fever on 8th day. Treated by udder inflation.

J.10 Friesian

5 years
Calved 23 August
Underfed 7-22nd day. post calving inclusive.
Stiff-legged gait on 20th and 21st days.

J.12 Friesian

5 years
Calved 24 August
Underfed 13-21st day post calving inclusive.
Stiff-legged gait on 19th and 20th days.

The other three underfed cows, and a fifth cow, put in as a replacement for J.8 to keep the feed ration constant for the others, all showed mild symptoms of tetany such as muscular tremors, stiff gait, knuckling at the fetlocks, and depression, but none went down.

The fattest cow of the group developed quite a strong ketosis in the second week of underfeeding, and lost about 300 lb. body weight, 200 lb. of which were not regained on re-feeding.

All cows, including the controls, showed hypomagnesaemia and several also showed hypocalcaemia, including a level of 4.9 mg.% in control cow E.3 two days after calving. She did not go down, although she appeared likely to do so several times. As stated above, the controls were rather underfed owing to lack of grass, but their hypomagnesaemia was much less marked and of a shorter duration than that of the underfed cows.

Discussion

During the course of the underfeeding experiments, a lowered serum calcium level was observed in some animals, as well as a lowered serum magnesium level. The cause of this hypocalcaemia is not fully explained, but Borgman and Reinecke (1950) have shown that thyroprotein antagonizes the parathyroid and may thus produce tetany in dogs. It is well known that sudden changes of feed, especially to fresh pasture or green cereals, and also periods of starvation, are liable to produce hypocalcaemia. The pregnant and the recently parturient ruminant appears to be especially susceptible to upsets of this nature.

While the administration of magnesium salts can lead to a temporary elevation of blood magnesium, we have found no method of artificially lowering it in cows other than by underfeeding after calving. Magnesium is not an element susceptible to *in vivo* interference by chemical means, such as complex formation, without first affecting the blood calcium.

The fact that it has been possible experimentally to lower the serum magnesium level is of great significance, because a low serum magnesium level is always found in clinical grass staggers.

The underfeeding and thyroprotein dosing experiments reported here have shown that deficient food-energy intake is one cause of hypomagnesaemia in lactating cows. Clinical cases of grass staggers have been produced by underfeeding alone, but with greater ease when thyroprotein dosing has been superimposed on the underfeeding.

Attempts to correlate the level of serum alkaline phosphatase (an enzyme activated by magnesium ions) with the magnesium level or with the onset of clinical grass staggers, have not been particularly successful. The normal range of serum alkaline phosphatase values in cows is very wide (Allcroft and Folley 1941), although normal individuals do not have wide day-to-day variations. Without preliminary normal samples, the results from field cases of metabolic disorders would have no value. In the underfed cows shown in Table 5 there appears to be some correlation between a fall in phosphatase level and underfeeding, and also with a fall in serum magnesium.

One of the problems connected with clinical grass staggers in this country has been the occurrence of a hypomagnesaemia in animals

on an adequate daily intake of magnesium. The tetany-paresis syndrome occurs in New Zealand on pastures in which there is no deficiency of magnesium (Cunningham 1936), and the hypomagnesaemia produced in the present experiments is not due to a deficient intake of magnesium.

Similar results on the effect of underfeeding and the development of hypomagnesaemia in cows have been reported by others. Breirem (1947) reported the production of tetany-paresis in cows by feeding rations twenty per cent below the standard energy content but containing sufficient protein in the form of herring meal. Ender, Halse, and Slagsvold (1948, 1949) produced hypomagnesaemia and slight ketosis with one fatal case of tetany and three of latent tetany by sudden reduction of the energy content of the ration of eight stall-fed cows. The protein intake was not correspondingly reduced, and was in excess of requirements as soon as body weight and milk production began to fall. The diets used were low in magnesium, and they found that the addition of magnesium to the diet protected their animals from hypomagnesaemia. Breirem, Ender, Halse, and Slagsvold (1949, 1954) note a marked individual variation in the serum magnesium level of cows on the same feed, and an individual predisposition to hypomagnesaemia. They conclude that an energy shortage is not the sole cause of the metabolic disorders, and interpret a low serum magnesium as being a sign of lability.

The fact that grass staggers sometimes occurs in association with a mild to moderate ketosis, and that, under the conditions of our experiments, the hypomagnesaemia was generally accompanied by a comparatively mild ketosis, indicates that a state of deficient food energy intake can occur in practice. Ketosis is, by definition, an excessive accumulation of ketone bodies in the animal organism; the result of catabolism, in the glycogen-deficient liver, of body-fat reserves to acetone bodies at a rate greater than that at which these substances can be utilized by the peripheral tissues. It follows that, for this condition to develop, the animal's energy requirement must be in excess of its food energy intake, and hence that it is underfed.

Underfeeding leading to hypomagnesaemia and grass tetany may result from sudden changes of feed in several ways. The change may be a reduction in the food and energy intake—absolute underfeeding. A change to a protein-rich pasture may, by adversely affecting the prevailing rumen flora, cause a digestive upset with consequent reduction in food intake or absorption—relative underfeeding; or, by the specific dynamic action of the protein-rich pasture, may produce a rise in metabolic rate and increased milk-energy output to give a negative energy balance—physiological underfeeding. This latter effect has been simulated experimentally, by dosing iodinated casein to give an increased metabolic rate and increased milk-energy output. The two factors, relative and physiological underfeeding, may operate together or in succession. This concept of "physiological" underfeeding, due to Eckles and Palmer (1916) has been confirmed by Graves (1947) who showed that high-producing mature dairy cows, even when fed ten per cent in excess of the calculated maintenance and production requirements, lost up to 100 lb. bodyweight in the early weeks of lactation. Woodward, Shepherd, and Graves (1933) also showed that fat cows at calving produced more milk and butterfat in the first part of the lactation period than thin cows; and that when fat they lost weight and when thin they gained weight

in early lactation. A similar result was obtained by Lees, McMeekan, and Wallace (1948) at Ruakura under conditions of good pasture grazing in early lactation. There was also a higher incidence of milk fever and grass staggers in the group that was well fed before calving, although both groups were grazed together after calving.

Some observations have been made on calving, oestrus, and weather; factors that could accentuate the effects of underfeeding on lactating cows.

The effect of calving was observed on two cows in mid summer. Grazing, ruminating, and lying-down times were all markedly reduced on the day of calving, while grazing time was subnormal up to the third day after calving. Serum magnesium was lowered slightly in the cow that took longer to return to normal grazing.

Oestrus is commonly accompanied by sudden changes in milk production, especially in highly strung cows, as well as by the increased physical activity and interference with grazing time resulting from bulling. The effect of oestrus on cows past the third month of lactation varied according to the intensity and duration of homosexual activity. Cows showing slight symptoms were unaffected. Those with marked sexual excitement and attraction grazed up to 50% less, lay down up to 60% less, showed a higher body temperature, a lower milk yield, higher fat content in the milk, and higher butterfat yield, and lost up to 10% in body weight on the day or days of oestrus as compared with the days before and after. The most active cow of a group of eight watched cows on which blood chemical studies were made, showed the only significant blood changes observed. Her serum magnesium and calcium content on the days before, during, and after oestrus were: 2.7, 2.0 2.7, and 10.5, 10.0, 10.4 mg.% respectively. A fall in the serum calcium at the time of oestrus has been common in our animals. No evidence has been found in the literature that oestrogenic hormones cause a fall in serum calcium in lactating cows. It may be assumed in the absence of such evidence that this occurrence is also a result of the physical disturbances of oestrus.

The effect of weather has been difficult to observe experimentally, but incomplete data have been obtained which suggest that very strong winds may cause a lowering of serum magnesium—a fall in four recently calved cows from 2.1 to 1.6 mg.% after a day on which 250 miles wind run was recorded on the anemometer. How this effect might occur is not known, but general observations indicate that strong wind interferes more with lying down than grazing, thereby possibly increasing grazing at the expense of rumination. This would be more likely to be of importance at night, when most rumination is done, and may lead to digestive upsets due to incomplete remastication of the fodder.

Limited study of the effects of a period of four days' heavy rain and strong wind on eight cows in mid- to late-lactation showed that these conditions were well tolerated by such cows, at least when fed liberal amounts of hay and green crops, even when little grass was obtainable. The only significant effect observed was a mean decrease in belly girth of three inches. Body weight was not determined.

Allcroft (1947) reported that the incidence of grass staggers was higher than usual in England in a particularly bad winter. The average

serum magnesium level in a herd of beef cows which he was studying fell very low at the beginning of this winter, but when hay and cabbages were fed out the magnesium level rose to normal, only to fall again when supplementary feeding ceased.

A spell of bad weather may either play a contributory part or may act as a precipitating factor in a number of cases which may have been just subclinical. Pastures become muddy and more feed is trampled and wasted than in fine weather. Hay becomes soaked and may be unpalatable, or worse still, the weather conditions may discourage the farmer from feeding out any at all. Energy losses may be increased by the cold, rain, and wind; and energy utilization increased by the lanes, gateways, and slopes becoming muddy, boggy, and slippery.

Thus it may be postulated that underfeeding in varying degrees of severity does occur in practice, and evidence has been obtained that this predisposes the lactating cow to a lowered serum magnesium level. Hypomagnesaemia and ketosis are not in themselves diseases, but biochemical abnormalities. The fact that it is possible to induce a severe hypomagnesaemia in an animal without its showing clinical signs of tetany would indicate that although hypomagnesaemia is a necessary condition for the tetany-paresis syndrome, it is not a sufficient condition. It has yet to be demonstrated what other factor causes animals with these abnormalities to develop clinically manifest disease. These diseases occur under widely different feeding and husbandry practices; and transit tetany, which resembles grass staggers clinically and biochemically, occurs when all food is withheld.

Our present knowledge, although still a long way from reaching a full understanding of the pathogenesis of these disorders, is sufficient to point to some of the conditions leading to grass staggers and ketosis, and so to methods of controlling them. As has been shown (Swan and Jamieson 1956b), modification in methods of herd management can, by avoiding these conditions, lead to a lowered incidence of disease. These conditions can also be used to provide susceptible animals for further investigation of the precise causes of clinically manifest hypomagenesaemia.

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THE DISTRIBUTION OF FERTILIZER FROM A BEAVER AIRCRAFT

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Summary

Two trials were conducted at Flock House in which measurements were made of the spread of fertilizers of differing particle sizes from a De Havilland Beaver aircraft. Under conditions of light wind a range of granular materials gave band-type distributions with effective widths of about 1 chain, the width increasing with a decrease in granule size. Super compound gave a less regular spread with an average width of about 90 ft. These fertilizers were dropped from a height of 200 ft. From a height of 400 ft. with suitable conditions and particle size, an increase in spread up to 2 chains was obtained in one drop. From a height of 100 ft. the spread was about 30 to 40 ft. and apparently was not dependent on particle size.

There was considerable loss of material fine enough to pass a 60-mesh sieve, but the extent of this loss with different wind conditions and flying heights was not determined.

Momentary changes in the strength and direction of the wind can cause large differences in the spread of most of these materials, and the technique of such trials must be improved to obtain results of sufficient accuracy to be of use in predicting the distribution patterns likely to be achieved in commercial operations under given conditions.

Introduction

Measurements of the spread of fertilizer from aircraft have been reported in this country by Campbell[1, 2], Miller[3] and Lynch[4], for aircraft of the Grumman Avenger and Tiger Moth (D.H.82) types. The spread from the De Havilland Beaver was measured in two trials conducted at the Flock House Farm of Instruction, Bulls, with a range of fertilizers of differing particle size. This machine is in commercial use, and its normal payload is 15 cwt.

The trials were carried out in December 1951 and March 1953 and in each case the field on which the drop was made was flat and was larger than the area required for measurement.

Details of Trial 1, December 1951

Five types of superphosphate were compared in this trial, all being dropped from a standard height of 200 ft.:

A. Aerial mixture No. 1 (Fine granulated)

B. Aerial mixture No. 2 (Medium granulated)

C. Aerial mixture No. 3 (Large granulated)

D. Granulated superphosphate ex England

E. Commercial "super compound".

The mechanical analysis of these fertilizers is shown in Table 1. Production of the aerial mixtures was arranged by Mr. M. M. Wright of the N.Z. Farmers Fertilizer Co. Ltd., New Plymouth.

TABLE 1. MECHANICAL ANALYSIS OF FERTILIZERS DROPPED. TRIAL 1

	Percentage Retained on B/S Sieve of Mesh				% Passed 60 Sieve
	8	16	30	60	
A. Aerial No. 1	2	51	31	13	3
B. Aerial No. 2	83	17	Tr	Tr	Tr
C. Aerial No. 3	100	Tr	Nil	Nil	Nil
D. Granulated super.	98	2	Tr	Tr	Tr
E. Super. compound	10	12	23	26	29

The plane made 3 runs for each fertilizer except the aerial mixture No. 3 for which there was only sufficient material for 2 runs. The load for each run was 7 cwt., and the hopper was kept open till it was empty.

The weather was fine with a breeze of from 7 to 9 knots in a direction such that most runs were made almost directly into it. However, it was by no means constant from moment to moment in direction of spread, and changed towards the end of the trial.

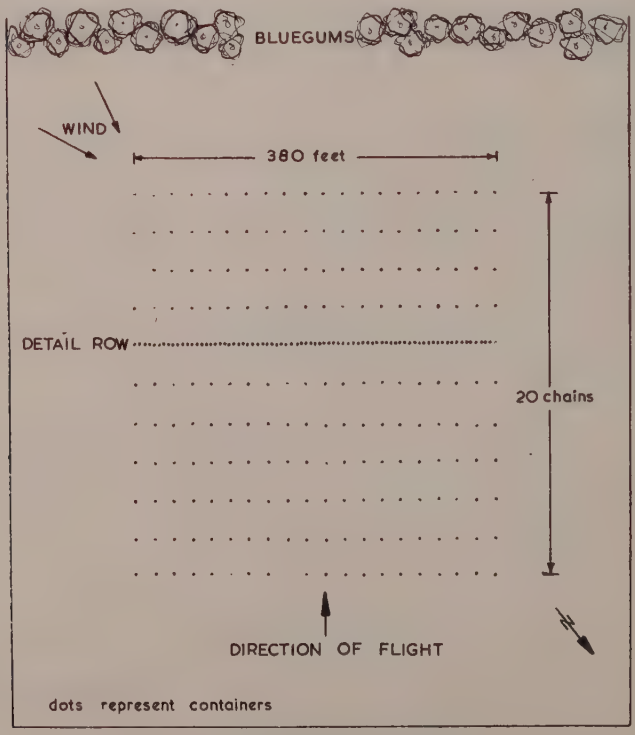

Fig. 1.—General plan of area of Trial 1.

The collecting receptacles were cardboard boxes 9 x 5 in. in area and 9 in. deep. For the super compound the boxes were lined with wax paper, so that fine dusty material was not lost. These boxes were set out in eleven rows at two-chain intervals, each row consisting of 20 boxes at 20 ft. intervals. The "rows" ran at right angles to the direction of flight. In one of the rows additional boxes were placed at 5 ft. intervals (Fig. 1).

The indicated air speed of the plane was 125 miles per hour during the trial.

Pattern of Spread

All the granulated materials gave clearly defined band patterns of distribution, though in some cases it was apparent that two lines of flight were so close together that the spreads from the two runs overlapped. This usually resulted in the spread from the other run for the same fertilizer being separated by a considerable gap of nil density.

The aim of the pilot was to achieve an even coverage of the area, but as granulated materials had not generally been available, the rate of flow and type of spread to be expected was not known to him, and even coverage was not achieved. However, this allowed a study to be made of the distribution from "single runs" in most cases.

The spread for the super compound was much less regular, and the density tended to vary almost as much along the direction of flight as across it.

The average widths of the bands where the density, as estimated from the amounts caught in the boxes, was $\frac{1}{2}$ cwt. per acre or more are shown in Table 2. The greater the proportion of finer fractions in the fertilizer, the greater was the width of spread.

TABLE 2. AVERAGE WIDTH OF SWATH RECEIVING $\frac{1}{2}$ CWT. PER ACRE OR MORE. TRIAL 1

	Feet
A. Aerial No. 1	72
B. Aerial No. 2	66
C. Aerial No. 3	54
D. Granulated super.	66
E. Super. compound	90

From Table 3 and Fig. 2, which shows the areas estimated to receive the different rates of fertilizer, the following points may be noted:

(a) for the three "aerial mixtures" there was a considerable area receiving more than 4 cwt. per acre, and this area was greater with materials of finer granule size. For the "medium" and "fine" mixtures an appreciable part of the area received more than 6 cwt per acre.

TABLE 3. ACREAGE COVERED BY DIFFERENT DENSITIES OF FERTILIZER. TRIAL 1

Cwt. per Acre	Fertilizer				
	A	B	C*	D	E
½–1	1.00	1.22	0.86	1.45	2.52
1–2	1.17	1.41	1.51	1.55	3.10
2–3	1.08	1.13	1.28	1.51	1.02
3–4	0.97	0.95	0.99	0.92	0.26
Above 4	1.18	0.95	0.69	0.36	0.15

* Adjusted to be the equivalent of 3 runs.

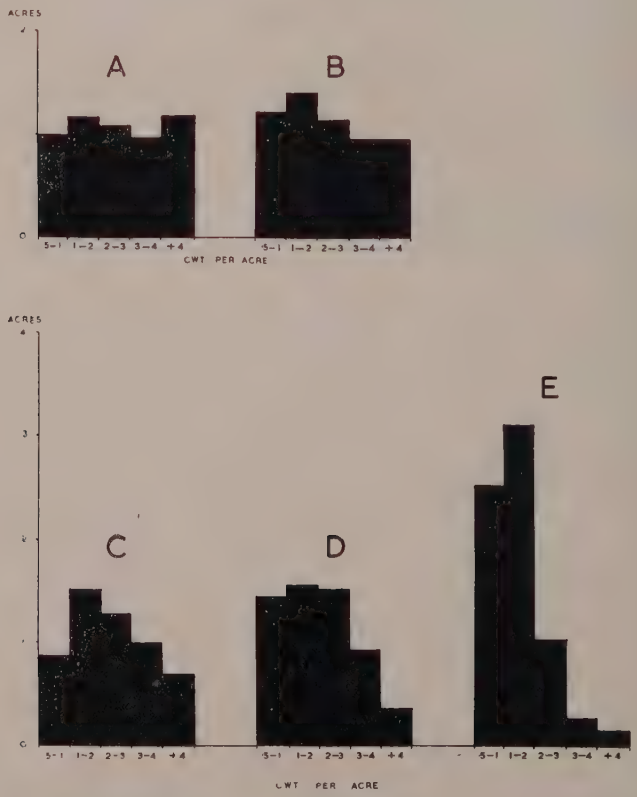

Fig. 2.—Block diagram of areas receiving different rates of each fertilizer.

(b) the area receiving between 3 and 4 cwt. per acre was equal for all granular materials.

(c) for all rates of coverage less than 3 cwt. per acre, the fertilizers of larger granule size showed greater areas covered. (Note that

the granule size of the English granulated fertilizer was slightly larger than that of the aerial mixture No. 3)

(d) the distribution pattern for the super compound was entirely different from that for any of the granulated materials, the largest area being that receiving $\frac{1}{2}$ to 2 cwt. per acre, with very little area receiving more than 3 cwt. per acre.

Thus, although the width of spread when taken to the $\frac{1}{2}$ cwt. per acre limit was greater with the finer granules, the peak density was higher and covered a larger area for these materials also. The length of swath varied considerably for different runs and, where this was short, one might expect greater densities to be recorded in cross lines. Examination of the graphs showed, however, that this was not the reason for the above effect, which was clearly demonstrated in average-length runs which could be studied singly, without any overlapping from adjacent runs. The percentage of the fertilizer dropped that could be accounted for by means of the sample was slightly higher for the aerial mixture No. 1 than for the other granular materials, but the difference was of a much smaller order than the differences in Table 3.

Separation of Particle Sizes

From the row with the boxes at 5-ft. intervals, more detailed graphs of the cross section of the distributions were drawn (Fig. 3). Because the pattern changes from point to point *along* the line of flight and because of the overlapping runs, these graphs by themselves do not give any information additional to that obtained above from the full grid. However, after weighing, the samples from these lines were bulked in threes and mechanically separated according to particle size. Results of this analysis are shown in Fig. 3. It is unfortunate that no separations were made of particles larger than those retained on a 8-mesh sieve. Hence the fertilizers in which the particles were nearly all of this size appear homogeneous in the graphs though they did in fact contain a range of particle sizes possibly as great as that of the other fertilizers.

No consistent sideways drift effect is apparent, though in some runs of the finer materials, such as aerial mixture No. 1 and super compound, the smaller fractions are slightly displaced relative to the larger fractions. The variable nature of this drift is probably a reflection of the changes in wind direction from moment to moment. From the second part of this trial it appears that the amount of this effect varies markedly with different heights of flying.

Recovery of Fertilizer Dropped

It has been suspected that a considerable amount of the finer fractions of powder materials such as superphosphate does not fall with the main bulk of the fertilizer but floats off in suspension. This loss may be greatly affected by speed of wind and its direction in relation to the direction of flying. One of the advantages claimed for granulated materials is the elimination of this loss and the more precise dropping of fertilizer on the target. Some effort was therefore put into obtaining reliable estimates of the "recovery" of each fertilizer from the target area.

From the box yields, maps were drawn showing contours of equal density of fertilizer. The areas between each pair of contour lines were measured and multiplied by the average density of the two lines. This

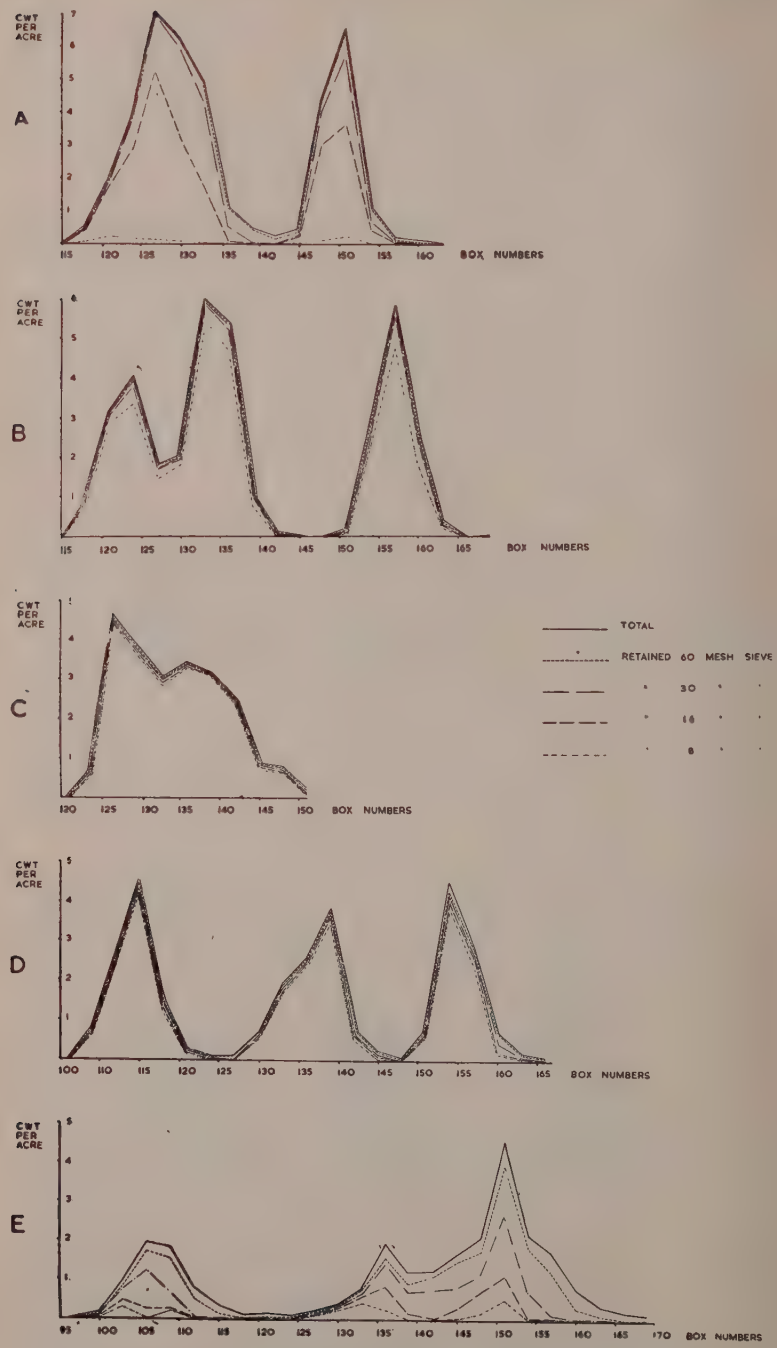

Fig. 3.—Cross sections of the distribution patterns, showing relative amount of different-sized particles, for all fertilizers in Trial 1.

computation gave the recoveries shown in Table 4. These are all surprisingly low.

TABLE 4. RECOVERY OF FERTILIZER. TRIAL 1

Fertilizer	Amount Dropped	Calculated Amount Falling on Area	% Recovery
	Cwt.	Cwt.	
A. Aerial No. 1	21	15.1	72
B. Aerial No. 2	21	14.4	68
C. Aerial No. 3	14	8.9	64
D. Granulated super.	21	13.2	63
E. Super. compound	21	10.8	51

It seems unlikely that any of the large granules would have become airborne and it is possible that they may have bounced out of the boxes. No completely satisfactory explanation of this loss can, however, be given. There is a slight trend in the granulated materials toward a higher recovery with smaller particle size, but the validity of this trend cannot be satisfactorily tested. However, in calculating these recoveries there were so many times when arbitrary decisions on boundaries had to be made that it is likely that the errors of the estimates are high and the trend may be quite fortuitous.

All errors in estimation, such as in transferring fertilizer from tins to bags or hence to the pans of the balance for weighing are naturally of a negative kind, but great care was taken to minimize these, and it is felt that they must be of negligible size.

The recovery of the super compound was fairly clearly lower than that of the granulated materials. With the "loss" of the granulated materials being unexplained, however, it seems difficult to assess how much of the super compound was actually "lost" off the target area by becoming airborne.

A different method of estimation was used on one of the fertilizers, granulated superphosphate (D), for which the spread pattern consisted of 3 separate and apparently uniform swathes. From the rows containing the closely spaced boxes, the average density per row was obtained and this was multiplied by the length of the swath. Again, arbitrary decisions had to be made, but the estimate by this method was close to that obtained by the contour-line method.

Effect of Height of Flying

A subsidiary trial was conducted later the same day to compare the spread obtained when dropping fertilizer from heights of 100 ft., 200 ft., and 400 ft. for super compound and for aerial mixture No. 1. A single row of collecting boxes placed at 10-ft. intervals across the line of flight was used, and the plane made one run at each height with each fertilizer.

The super compound at both 100 and 200 ft. showed a peak density of only about 2 cwt. per acre which was less than that generally recorded in the main trial, but the proportions of the different fractions recorded appeared similar to those recovered in the main trial. There was a sideways displacement, particularly of the smaller particles and this was similar for both heights (Fig. 4). At 400 ft. all the super compound was carried away sideways beyond the target area. The pilot endeavoured to make allowance for the sideways drift in setting his course, but in this case was apparently not successful. The actual path of the plane in relation to the position of the boxes was not charted.

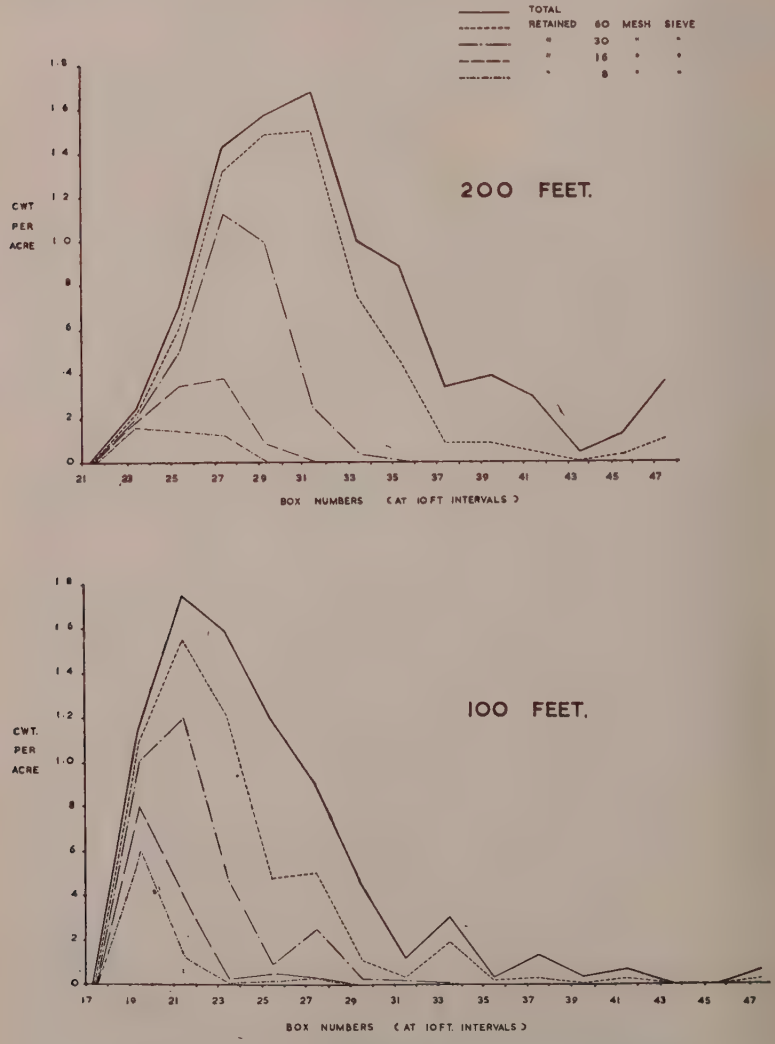

Fig. 4.—Cross sections of the distribution patterns of super. compound from heights of flying of 100 and 200 ft.

For the aerial mixture No. 1 (fine granulated), the peak density and the average rate were much higher for the 100 ft. run than for the 200 ft. run, although the particle-size distribution was similar for the two runs. Results from the main grid indicate that there can be considerable variation in the rate sown from point to point along a run and little significance should be attached to this difference, which may not have applied to the whole run. The width of swath to the $\frac{1}{2}$ cwt. per acre

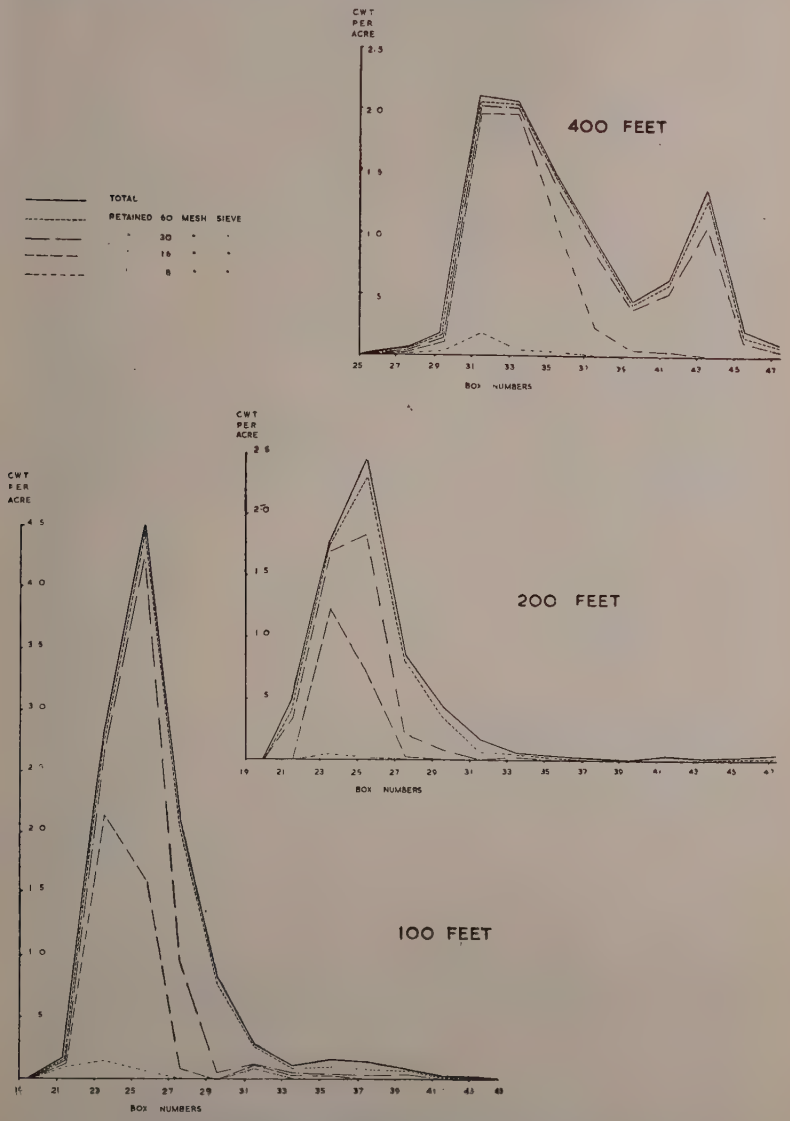

Fig. 5.—Cross sections of the distribution patterns of Aerial Mixture No. 1 from heights of flying of 100, 200, and 400 ft.

limits (as estimated from the amount actually caught in the boxes) was equal, but this may not be a fair comparison in view of the different mean rates recorded. There is evidence of a slight sideways displacement particularly of the finer particles from both heights (Fig. 5).

At 400 ft. the distribution curve showed a double peak, and the particle analysis showed that, whereas the first peak consisted mainly of material greater than 16 mesh, there was none of this size in the second peak which mainly consisted of the 16-30 mesh material. Although the density at the larger peak was only 2.2 cwt. per acre, which was equal to that for the 200 ft. run, the swath width to the $\frac{1}{2}$ cwt. per acre limit was 130 ft., compared with 78 ft. for the 200 ft. run.

It must be emphasized that all the results that have been quoted as rates per acre refer to the amounts collected in the boxes. Because the estimated recovery percentages were only of the order of 70% even in the best of the granular materials, the actual rate deposited per acre is probably higher than that estimated from the boxes, and the widths quoted to "$\frac{1}{2}$ cwt. limits" really refer to limits a little higher than $\frac{1}{2}$ cwt.

DETAILS OF TRIAL 2, MARCH 1953

The first trial suggested that the ideal material for dropping from this type of plane would be one with no "dust" fraction (passing a 60-mesh sieve) but with a range of particle sizes down to this limit. To obtain such a material, a quantity of serpentine superphosphate was sieved, and that portion passing a 16-mesh sieve but retained on a 60-mesh sieve was mixed with an equal quantity of aerial mixture No. 2. The sieved material alone was also dropped for comparison. Details of the fertilizers dropped are:

A Sieved serpentine superphosphate.

B 50:50 mixture of sieved material and Aerial No. 2.

C Aerial mixture No. 1.

D Commercial serpentine superphosphate.

E Super compound of the same line as used in the previous trial.

The mechanical analysis of the samples taken as the fertilizers were loaded into the plane is shown in Table 5.

TABLE 5. PARTICLE-SIZE DISTRIBUTION OF FERTILIZERS. TRIAL 2

	Percentage Retained on B/S Sieve				% Passed B/S Sieve
Fertilizer	8	16	30	60	60
A. Sieved serp. super.	—	5	40	46	9
B. 50:50 sieved super. and aerial No. 2	31	13	22	27	7
C. Aerial No. 1	5	47	29	15	4
D. Serpentine super.	2	4	23	32	39
E. Super. compound	10	10	18	24	35

TABLE 6. PERCENTAGES OF DIFFERENT PARTICLES RECOVERED FROM THE DROP. TRIAL 2

Fertilizer	Percentage Retained on B/S Sieve				% Passed B/S Sieve
	8	16	30	60	60
A. Sieved serp. super.	—	4	40	50	5
B. 50:50 sieved super. and aerial No. 2	47	12	21	17	3
C. Aerial No. 1	6	58	26	8	2
D. Serpentine super.	2	5	32	45	17
E. Super. compound	8	13	33	32	14

Conditions for this trial were unfortunately very poor. The wind quickly rose from 10 knots at the beginning of the trial up to 18 knots, and in order to ensure that the fertilizer would land over the grid of boxes the pilot considered it necessary to fly at 100 ft. His actual height may in fact have been lower. Details of the area are shown in Fig. 6.

To overcome the loss due to bouncing of the granules, the collecting receptacles used were kerosene tins 9 × 9 in. in area and $13\frac{1}{2}$ in. high. A slightly different grid was used, the aim being to get accurate pictures of the crosswise distribution at three selected places and also an accurate measure of the total length of each run. Three runs were made with each fertilizer except C and E for which supplies were short and only two runs could be made. The aim of the drop was to have clearly separated swaths for each run so that widths of spread could be compared, and this was achieved except for fertilizer A where the last two runs overlapped, and unfortunately the first run was faulty because of a leaking hopper.

Pattern of Spread

Because of the low height at which the pilot flew in the rather strong wind, the width of spread of all fertilizers was very small and it was impossible to detect differences in width of spread with boxes spaced at 10 ft. intervals. Also the length of spread varied considerably with the different materials, and this resulted in different rates being dropped over the transverse measuring lines. With differing rates, a comparison of the widths to an arbitrary figure such as $\frac{1}{2}$ cwt. is not a valid estimate of relative efficiencies of spread.

The rate applied seems to vary considerably from one cross line to the next for a number of the materials—although again the boxes were not close enough to measure this accurately.

Separation of Particle Sizes

The graphs of cross sections of the spread were symmetrical for all materials, both in total amounts and in regard to the different fractions in the material. This indicates that there was no spreading out of fertilizer to one side. The wind was in general nearly parallel to the

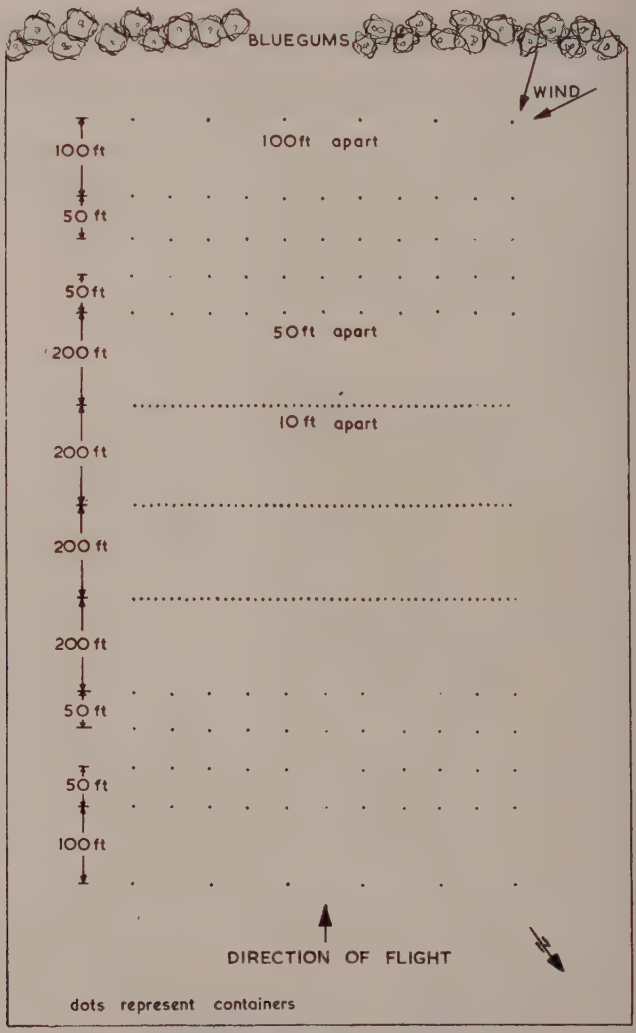

Fig. 6.—General plan of area of Trial 2.

direction of flying. It was apparent however that the serpentine superposphate and super compound had a much smaller proportion of the fine material in the collecting tins than had been in the original material. A comparison of Tables 5 and 6 shows that the percentage of material passing the 60-mesh sieve dropped from 39 to 17 for serpentine super and from 36 to 14 for super compound. For the sieved serpentine superphosphate the percentage of material between 30- and 60-mesh sieve dropped from 27 in the original fertilizer to 17 in the tins. A possible explanation for this is that the 30-60 fraction in this fertilizer was near the 60-mesh limit and also became airborne. Otherwise the relative proportions of the particles of different size were the same in the tins as in the original material.

Recovery of Fertilizer Applied

The tins were arranged to give a detailed picture of the crosswise spread on 3 lines and also to indicate accurately where the beginning and end points of each run were. However, many runs were not long enough for the end points to be among the cluster of tins set to measure this, and thus the lengths of the runs could not be determined, and the recovery could not in general be assessed. Two runs of fertilizers A (sieved serpentine superphosphate) and D (serpentine superphosphate) did appear satisfactory for estimation. For D, the amounts recovered from each of the 3 crosswise lines were similar, and on this basis the recovery was estimated as 54%. For A, however, the amounts collected in the 3 cross lines were in the ratio of 41, 59, and 90 respectively. This makes the use of an average figure for the crosswise spread somewhat doubtful, but the recovery figure using an average was 73%. For this sieved serpentine superphosphate, the material recovered showed the same particle distribution as the original sample, and observations during the trial indicated that bouncing out of the kerosene tins was not taking place, so that the loss of 27% is unexplained except as the result of an inadequate sample or an incorrect method of estimating the recovery from the given sample. This is a serious flaw in the technique, and a comparison of the catches in tins with some kind of measurement from a continuous strip across the target area would seem desirable before further trials are undertaken.

DISCUSSION

In spite of great care in planning and much work in the execution of trials of this kind, it is difficult to be sure of obtaining detailed and consistent information when the trial must be done on one specified day regardless of wind conditions, unless the wind is too strong for safe flying.

In comparing fertilizers, it would be desirable to know the exact rate of discharge of each over the target area. Although the weather conditions cannot be controlled in commercial operations, it is desirable that variability due to them be "controlled" in experimental work. All comparisons should ideally be made under a variety of wind speeds.

The findings of these two experiments with the distribution of fertilizer from a Beaver aircraft may be summarized as follows:

Technique

The techniques used of having about 300 small receptacles spread in some kind of grid throughout the area of deposition of the fertilizer have not proved entirely satisfactory. Not more than 70% of the fertilizer dropped has been accounted for, even when the material consisted almost entirely of large granules. Because it is difficult to specify beforehand what will be the probable length and width of spread which will be obtained, many more boxes than 300 seem necessary to ensure that a reliable picture will be obtained for most runs of the aircraft, unless the boxes are intended to cover only the area of deposition from single runs. The more meteorological records that can be obtained, particularly of wind speeds and directions, during the actual dropping of each fertilizer, the better.

Loss of Fertilizer

Under most conditions there is considerable loss through wind drift of material finer than 60-mesh, when dropped from heights of

100 to 200 ft. Loss of material larger than this has not been satisfactorily assessed.

Pattern of Spread

At 200 ft. height of flying under low wind conditions, the width of spread is increased with decrease in particle size, but details of this relationship have not been accurately ascertained. At 100 ft. height, particle size, in fertilizers that are almost wholly granular, does not play an important part. At 400 ft., if conditions are such that the material can be dropped accurately over the target area, there are indications that an ideal fertilizer would be one containing a mixture of particles of sizes between 8- and 60-mesh sieves and that a spread of up to 130 ft. could be obtained. At this height, however, a slight increase in wind speed could carry almost all the material slightly off the target area. No details of the operation of this effect are available from these trials. At 200 ft. the width of spread to limits of $\frac{1}{2}$ cwt. with an attempted rate of 3 cwt./acre of granular materials was about 1 chain, and at 100 ft. or less this was reduced to about 30 to 40 ft.

Acknowledgements

In these large experiments, much help and co-operation was received from the staff and trainees of Flock House, the R.N.Z.A.F. Station at Ohakea, the pilots and staff of Rural Aviation Ltd., from Rukuhia Soil Research Station for the mechanical analysis of all the samples, and from all staff of the Crop Experimentalist's Section.

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THE EFFECT OF MCPB (SODIUM 2-METHYL-4-CHLOROPHENOXYBUTYRATE) ON YOUNG RED CLOVER (*Trifolium pratense*) AND WEEDS

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Summary

A weedy establishing red-clover sward was sprayed with MCPB (sodium salt) at rates of 0, 1, 2, 4, and 6 lb. acid equivalent per acre and with MCPA (sodium salt) at 1 lb. acid equivalent per acre. The effects of these treatments, on yield and growth form of clover and weeds, were studied.

MCPB did not depress red clover yield significantly, but leaf scorch and distortion of petioles and of later leaves was produced by heavy rates of the material. Red-clover yield was reduced by MCPA. Weeds were reduced by both materials.

MCPB was slower in action and less effective than equivalent MCPA. From observation, 1 lb. MCPA was about equal to 2 lb. MCPB in its effect on weeds. Weeds observed to be susceptible or not are listed. Docks (*Rumex crispus* and *R. obtusifolius*) were susceptible until the flowering stage.

Introduction

Weedkillers in current use, notably the selective "hormone" weedkillers generally cause damage to clovers (Edmond 1955, Scragg 1953, Holly 1953, Ochiltree 1954) and this restricts their use on pastures. A new approach to selective weed control has been made possible by the discovery that some plants, by a β oxidation mechanism, are able to oxidize some of the higher homologues of ω phenoxyalkylcarboxylic acids to the physiologically active acetic acid homologue (Wain 1955). Already it is known that a number of plants are not activators of these acids (Wain 1954a), and it appears that both white and red clovers fall within this group (Wain 1954b). Should this be correct, there would be no need to restrict application rates in order to avoid damaging clovers.

It was decided to determine the effects of heavy rates of 2-methyl-4-chlorophenoxybutyric acid, as the sodium salt (MCPB) on an association of young red clover and weeds.

Materials and Methods

On 17 September 1955, the experiment area was sown broadcast with red clover at 12 lb. per acre. The soil (Manawatu sandy loam) is light and shallow, overlying gravel, and readily dries out in a normal summer. Many weeds established, and in November the field was mown twice. After the clover had reached a height of 4 in. following the second mowing, spray treatments were applied on 9 December 1955 through low-volume equipment at the rate of 20 gallons of solution per acre and 30 lb. per square inch pressure. On a 6 × 6 latin square, each plot was 13 ft. wide and 65 ft. long.

The treatments were:

1. No treatment—control.
2. Sodium salt of MCPA (30% acid equivalent) at the rate of 1 lb. acid/acre

3. Sodium salt of MCPB (40% acid equivalent) at the rate of 1 lb. acid/acre
4. Sodium salt of MCPB (40% acid equivalent) at the rate of 2 lb. acid/acre
5. Sodium salt of MCPB (40% acid equivalent) at the rate of 4 lb. acid/acre
6. Sodium salt of MCPB (40% acid equivalent) at the rate of 6 lb. acid/acre.

The visible effects of treatments and the plot yields were recorded on 20 and 29 December 1955 and 4 and 11 January 1956. At each harvest, two strips each three feet wide were cut with a sickle-bar mower across each plot at randomly chosen sites, and green weight, oven-dry weight, and botanical composition of the herbage were determined.

Red-clover seed which set in late January was harvested by hand from all plots in late February and tested for germination at 20°C.

When the trial began, the distribution of red clover and weeds appeared to be uniform but because of later differential drought effects this uniformity was lost.

RESULTS

Total Herbage Yield

The differences in yield between treatments were highly significant at each harvest and did not change significantly (Table 1). The reduction in yield in sprayed treatments occurred mainly in the weed species.

TABLE 1. MEAN DRY-WEIGHT YIELDS (g./sq. yd.) FOR THREE SPECIES CLASSES AND RESULTS OF ANALYSES OF VARIANCE

Treatment	All Species	Red Clover	Weed Species
Control	154.6	47.7	90.4
MCPA 1 lb.	78.6	23.3	34.5
MCPB 1 lb.	94.9	35.3	39.4
MCPB 2 lb.	96.0	44.4	31.9
MCPB 4 lb.	89.0	42.4	28.6
MCPB 6 lb.	83.3	41.1	27.0
Standard error	± 6.1	± 4.1	± 3.8
Sig. diff. at 5%	17.9	11.5	11.1
F for treatment	21.16**	4.65**	41.41**
F for interaction (Tr x Cut)	1.32n.s.	1.21n.s.	2.63**
Coefficient of variation	29%	48%	34%

Note: All-species data derived from four harvests, red clover and weed species from three.

Red Clover

The red-clover yield was significantly reduced only by MCPA.

It was observed that, within 24 hours of spraying, MCPA caused severe distortion of petioles and wilting of leaves, but MCPB at the same rate had no discernible effect. At the higher rates of MCPB the upper leaves in particular showed signs of spray scorch, the effect being slight at the 2 lb. rate and pronounced at the 4 and 6 lb. rates. Two days after spraying when the scorch effect had appeared, sodium chloride

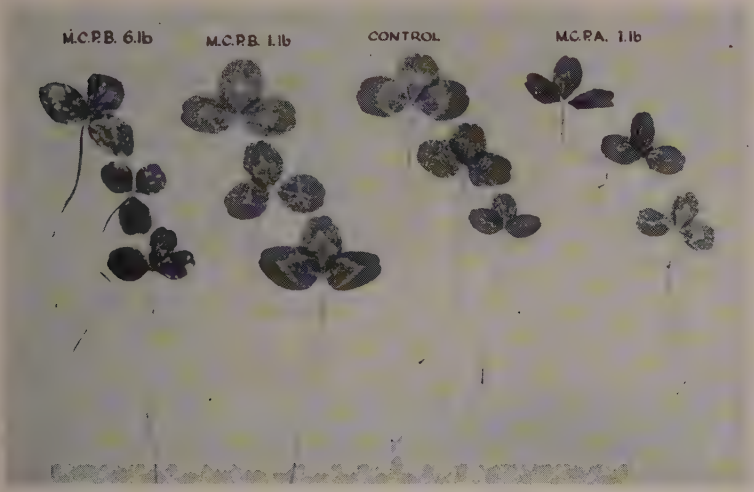

Fig. 1.—Red-clover leaves and petioles distorted by MCPA, and scorched and distorted by MCPB.

Fig. 2.—Delayed effect of MCPB on red clover shown by distorted frond-like leaves; earlier and later leaves are normal.
Left: MCPB. *Right:* Untreated.

was applied to the surrounding sward at the same dilution as the 6 lb. MCPB rates. The resultant scorching was less severe than with MCPB. MCPB produced distortion of petioles at the 4 lb. and 6 lb. rates, but it was not severe (Fig. 1).

A further feature was that the new leaves which appeared in MCPB plots for a short period commencing eight to ten days after spraying were distorted. The same distortion was observed in MCPA plots commencing ten to fourteen days later. In each case leaves which appeared later were normal (Fig. 2).

Weed Species

Weed species yield was reduced significantly by all spray treatments (Table 1). The highly significant Harvest x Treatment interaction shows that the treatment effect changed with time, the differences between treatments becoming greater (Table 2).

TABLE 2. Weed Species: Treatment Means for Harvest x Treatment Interaction (g/sq. yd.)

Treatment	Harvest Dates		
	20.12.55	4.1.56	11.1.56
Control	76.0	100.2	95.2
MCPA 1 lb.	49.3	36.3	17.8
MCPB 1 lb.	44.2	45.5	28.5
MCPB 2 lb.	40.8	29.5	25.3
MCPB 4 lb.	39.3	31.5	14.8
MCPB 6 lb.	34.5	27.8	18.7

Standard error ± 5.8 Sig. diff. at 5% = 17.2

At the first harvest there was a significant relationship between increased rate of MCPB and reduction in weed yield.

Both MCPA and MCPB began to take effect with equal speed, but later the MCPB effect appeared slower and less severe in action. Observation suggested that 2 lb. MCPB were about as effective as 1 lb. MCPA. An explanation for the fact that the yield data do not support this observation is that the dry soil conditions reduced the continued growth of unaffected plants and prevented decay of dead plants. The following species were observed to be activators:—

Capsella bursa-pastoris
Chenopodium album
Coronopus didymus
Plantago lanceolata
Plantago major
Polygonum persicaria
Rumex crispus
Rumex obtusifolius
Sisymbrium officinale
Solanum nigrum

However only in three of these were yields significantly affected on 4 January 1956 (Table 3).

TABLE 3. MEAN DRY-WEIGHT YIELDS (g./sq. yd.) FOR THREE SPECIES AND RESULTS OF ANALYSES OF VARIANCE, 4.1.56

Treatment	*Rumex* spp.	*Sisymbrium officinale*	*Polygonum persicaria*
Control	3.20	3.21	5.85
MCPA 1 lb.	1.80	1.72	3.33
MCPB 1 lb.	1.12	1.26	3.83
MCPB 2 lb.	1.12	1.29	2.97
MCPB 4 lb.	1.80	1.50	2.33
MCPB 6 lb.	1.39	1.46	2.92
Standard error	± 0.34	± 0.46	± 0.36
Sig. diff. at 5%	1.01	1.34	1.06
F for treatment	5.17**	2.65(*)	11.77**
Coefficient of variation	48%	64%	25%

Note: Analyses and results based on transformation $\sqrt{1 + x}$

Species which did not appear to be activators:—

Conium maculatum — *Silene gallica*
Matricaria spp. — *Spergula arvensis*

The effect on flowering docks (*Rumex crispus* and *R. obtusifolius*) and *Polygonum persicaria* became greater with time; that on *Sisymbrium officinale* was not great, because of natural death of plants.

Seed

There were no treatment effects on the germination of red-clover seed, all samples when scarified giving a germination in excess of 90%.

DISCUSSION AND CONCLUSIONS

MCPB did not reduce the total yield of red clover significantly, but, on 11 January, the untreated clover appeared more vigorous than the rest. Drought may have caused this, by greater wilting of clover, in sprayed areas. It is most unlikely that MCPB effects on red clover were responsible for this difference.

Of particular interest is the observation that many docks were killed by the MCPB. It may thus be possible to check this perennial weed in pastures, even at the flowering stage.

The general effectiveness of 1 lb. MCPA appeared to be more closely approached by 2 lb. MCPB than by 1 lb., and two weeds at least (*Polygonum persicaria* and *Coronopus didymus*) were affected more slowly by MCPB than by MCPA. The full significance of these observations is not clear.

Species varied in susceptibility to MCPB, as did variously aged plants of any one species. Consequently, the mature state of most weeds in this trial probably restricted the effects of the spray treatments.

These results agree with those of Wain (1954a, b) and Carpenter and Soundy (1954) but the leaf scorch, petiole distortion, and delayed leaf distortion noted were not reported by them. It is possible that these were not observed at the low rates of MCPB they used. The adverse effect of MCPA on red clover (Edmond 1955, Scragg 1953, Holly 1953, Ochiltree 1954) is confirmed, which serves to stress the advantage to be gained by using MCPB. A number of the weed species observed to be susceptible to MCPB have previously been reported to be activators (Wain 1954b, Carpenter and Soundy 1954) but it has not been recorded that the susceptibility of *Rumex* spp. persists into the flowering stage.

It may be concluded that, at the 1 lb. rate, MCPB does not harm red clover, but it does suppress susceptible weeds.

Acknowledgements

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RUMEN STUDIES IN SHEEP

III. THE EFFECT OF FEED INTAKE ON THE CONCENTRATIONS OF MICROBIAL END-PRODUCTS

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(Received for publication, 14 May 1956)

Summary

1. Twelve mature sheep were fed dried grass of uniform composition, the ration being decreased at intervals from 1,000 to 400 g/day. Rumen samples were taken at each feed level.

2. A highly significant positive correlation of ruminal volatile fatty acid level with intake was found. The acetic/propionic ratio was significantly higher at the 400 than at the 1000g/day level. Ruminal pH, ammonium nitrogen, protein nitrogen and free microbial count were unaffected by change in intake.

3. The dry matter content of the rumen ingesta of 32 sheep was found to vary between 3.8 and 7.5 per cent; the implications of this finding are briefly discussed.

Introduction

In a field trial carried out earlier[1], differences in concentrations of rumen microbial end-products were obtained in two groups of sheep on different pasture treatments. Since the herbage intake of one group was only 60% of that of the other, it seemed possible that this might have accounted for the differences found in the rumen.

So far, microbial end-products and free population have been determined as concentrations only, and it was considered that these should reflect their rates of production. The rate of production per unit volume is, however, a direct function of two variables: microbial activity per unit feed, and feed per unit volume. If feed per unit volume is reduced by decreased feed intake, concentrations will be lowered and no longer represent activity per unit feed, which is the true criterion of rumen function. Little information is available on the variability of feed per unit volume, though calculation from the limited data of Balch *et al.*[2] suggests that the dry matter content of the rumen is fairly constant.

It was therefore decided to test whether rumen concentrations could be altered by varying the intake of a standard high-quality feed, and for this purpose dried grass was selected. Information was also sought on the constancy of feed per unit volume.

Experimental

Twelve mature wethers were individually penned and fed dried grass which had been cut from spring pasture consisting mainly of perennial ryegrass, white clover, and prairie grass, and dried for 30 minutes in a stream of air at 100–120°c. A feed level of 1,000 g/day was maintained for 5 weeks and then decreased to 400 g/day in steps of 100 or 200 g. Half the ration was fed at 0800 hours and half at 1500 hours. Feeds were always consumed within 30 minutes. Water was always available.

TABLE 1. Effect of Varying Feed Levels

Feed* Intake (g/day)	Live Weight (lb.)	Rumen Samples per Sheep	pH	Ammonium N. (mg. %)	Protein N. (mg. %)	Microbial Count (x 10^6/cu.mm.)	VFA (millimoles %)	VFA Percentage (millimoles %)	VFA Ratios (millimoles %)
1000 33 days	107 ± 8	5	6.5 ± 0.1	19.8 ± 3.5	165 ± 32	24 ± 5	9.7 ± 0.8	Ac.66.0 ± 1.8 Pr. 23.4 ± 1.1 Bu.10.6 ± 1.0	Ac./Pr.2.8 ± 0.2 Pr./Bu.2.2 ± 0.2 Ac./Bu.6.3 ± 0.8
800 5 days	105 ± 8	1	6.6 ± 0.1	22.7 ± 1.8	167 ± 38	—	8.9 ± 0.8		
700 5 days	102 ± 8	1	6.6 ± 0.1	24.9 ± 3.6	177 ± 43	—	8.1 ± 0.9		
600 12 days	102 ± 8	1	6.7 ± 0.1	19.3 ± 1.9	153 ± 30	—	7.3 ± 0.7		
500 8 days	99 ± 8	2	6.6 ± 0.1	20.0 ± 2.4	173 ± 32	26 ± 7	7.5 ± 0.8		
400 13 days	94 ± 7	3	6.6 ± 0.1	19.4 ± 3.1	161 ± 36	22 ± 5	7.1 ± 0.8	Ac.68.3 ± 1.7 Pr. 21.2 ± 1.0 Bu.10.5 ± 1.2	Ac./Pr.3.2 ± 0.2 Pr./Bu.2.0 ± 0.2 Ac./Bu.6.6 ± 0.8

* Approximately 85% dry matter.

Live weights were recorded weekly.

After a preliminary period of 2 weeks, rumen samples were taken weekly or twice weekly and analysed for pH, ammonium nitrogen, protein nitrogen (PN), volatile fatty acids (VFA), and free microbial count, using methods previously described[1]. Chromatographic separations of individual acids were carried out on the VFA distillates bulked for each sheep at the 1,000 and 400 g/day levels.

The rumen contents of 32 sheep were weighed at slaughter, after the animals had been starved for 18 hours, and samples were dried to constant weight at 105°C.

RESULTS

The data from the pen-feeding trial are summarized in Table 1. Differences in concentration due to intake level were found in VFA, which gave a highly significant positive linear correlation with intake (0.44 millimoles per cent per 100 g feed or 0.74 millimoles per cent per 100 g digestible organic matter.) The ratio of acetic to propionic acid was slightly but significantly higher at the 400 g level than at the 1,000 g level. The proportion of butyric to other acids showed no change. All other end-products measured were unaffected by feed level.

The rumen dry-matter contents of the 32 sheep examined showed a range of 3.8% to 7.5% with a standard deviation of $\pm$ 0.8%.

DISCUSSION

The effect of intake level on VFA without corresponding changes in other end-products is unexpected and not easy to explain. It might be suggested that different levels of intake brought about differential changes in the activity of the microflora towards the carbohydrate and nitrogenous constituents of the feed, but no effects of this nature have so far been reported.

If decrease in feed intake results in reduced feed per unit volume, a fall in the concentrations of all end-products would be expected. Feed per unit volume, as measured by the dry-matter content of the rumen, is seen from the results to be widely variable and this may account for the large variability found in concentrations[3]. The only explanation for the change in VFA alone appears to be a differential change in the rate of absorption of ammonia and VFA by the epithelium. This might be caused either by altered concentration gradients with the venous circulation or by modification in the selective permeability of the epithelium which is known to operate even against the concentration gradient[4]. Rumen epithelial tissue actively metabolizes VFA, but not ammonia, to provide the energy required for the transference of molecules and ions, including VFA[5], and this in itself may be an important consideration, particularly since the weight of tissue to be supplied remains the same even if the total production of VFA is decreased. Ruminal pH, which has been shown to have a profound effect on VFA absorption[6], did not change during this trial and hence could not have been a contributing factor.

Certain reservations must be made in applying these results to field experiments. It is not known whether the weight or condition of the animal plays any part in rumen concentrations, and in this trial it was

not possible to control these factors. Nor was it feasible to reproduce field conditions with regard to the dry-matter content of the feed, the rate of its ingestion or the amount of water drunk, all of which might have exerted some influence. Evidently the measurement of rumen volume by a technique such as that of Sperber *et al.*[7] would be of the utmost value in elucidating the effect of these factors on rumen activity.

It is interesting to note that the PN and free microbial count were much lower when the animals were fed dried grass than under grazing conditions. This suggests that the denaturation resulting from the drying process rendered the protein less available to the microflora, although the grass was highly digestible (78% organic matter) and of excellent fattening quality. Chalmers *et al.*[8] have shown that rumen fermentation of certain proteins is affected by previous heat treatment and further work is planned to investigate the effect on rumen activity of changes in herbage composition due to heat drying.

Despite the limitations of this trial, it is clear that in any rumen work involving VFA levels or proportions, intake levels must be taken into consideration and where possible standardized.

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OLEANDER KNOT
(*Pseudomonas savastanoi* (Erw. F. Smith) Stevens)

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(*Received for publication, 24 July 1956*)

Summary

Pseudomonas savastanoi is recorded for the first time in New Zealand, causing oleander knot on *Nerium oleander*. This organism also caused galls when inoculated into olive stems.

Disease symptoms, morphology, and cultural and physiological reactions of the causal organism are described.

Nomenclature of *Ps. savastanoi* is discussed.

Introduction

During 1955, specimens of oleander (*Nerium oleander* L.) infected with a gall disease on leaves and stems were received from Nelson, Hastings, and Wellington. The galls were found to be caused by bacteria and a study has been made of the strains isolated. Oleander knot is of little economic importance in New Zealand. The causal organism is, however, of scientific interest because of its similarity to other plant-gall-forming bacteria.

Symptoms

Oleander knot is characterized by the formation of galls or knots on stems, foliage, and inflorescence of oleander. On young shoots the disease may develop as a series of longitudinally arranged swellings (Fig. 2a) which split the epidermis to form a long canker or a number of galls. Greenish cream-coloured gall tissue continues to develop at the edges of the dark-brown to black canker (Figs. 1b and 2a). On older stems the galls in early stages are more or less spherical and corky, up to 2 cm. in diameter, with relatively smooth surfaces. They are at first coloured greenish cream but later the knots become irregular and rough, dark brown to black in colour with projecting tubercles (Fig. 1a). The bacteria appear to stimulate secondary cambium formation which gives rise to this corky tissue.

At first the leaf spots are small corky projections on either leaf surface, sometimes surrounded by a yellow halo, later becoming black and necrotic (Figs. 2b and 3). Deep pits often occur on the reverse side of leaves beneath infection, causing leaf distortion. Infection of the midrib is common, the localized galls being similar to those on stems but of smaller dimensions (Fig. 3), but frequently long cankers occur on midribs (Figs. 1a and 2b). If young leaves become infected as they

emerge from buds, they become distorted and thickened with inrolled margins.

In New Zealand, infected flowers or seed pods have not been observed, but Elliot (1951) describes the following symptoms. "Infected pistils are distorted and the petals remain attached for a long time. Young seed pods are shorter and thicker than normal and have aborted seeds. Seed pods infected when partly grown become curved and distorted but produce normal seed".

Fig. 1

Fig. 2

Fig. 1. a.—Advanced dark-brown canker with projecting tubercles caused by the oleander-knot bacterium. Natural infection.
b.—Longitudinal canker surrounded by actively growing gall tissue. Natural infection on oleander.
—*Photo: J. W. Endt.*

Fig. 2. a.—Typical longitudinal canker and small secondary tubercles on young oleander stem. Natural infection.
b.—Leaf infection (natural) showing typical midrib canker and localized lesions. Note faint yellow halo around small lesion at lower right.
—*Photo: J. W. Endt.*

Isolation and Pathogenicity of Organism

The bacterium responsible for this disease is readily isolated on beef peptone media from fresh gall tissue. To test pathogenicity many single colony isolations from leaf and stem galls were needle inoculated into leaves and stems of *Nerium oleander* plants. Typical galls developed (Fig. 3) from which the pathogen was re-isolated and used for further successful inoculations. The original isolates were also inoculated into stems of olive (*Olea europaea* L.) plants and corky, more or less spherical,

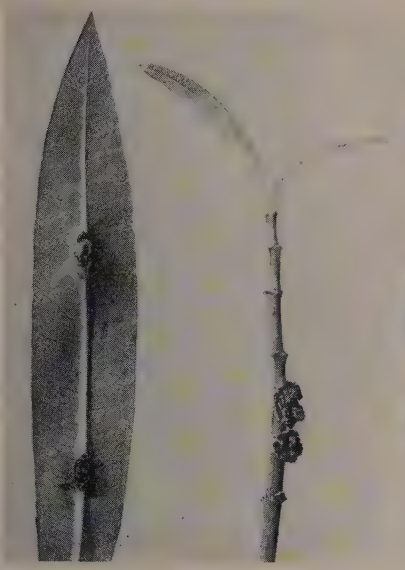

Fig. 3

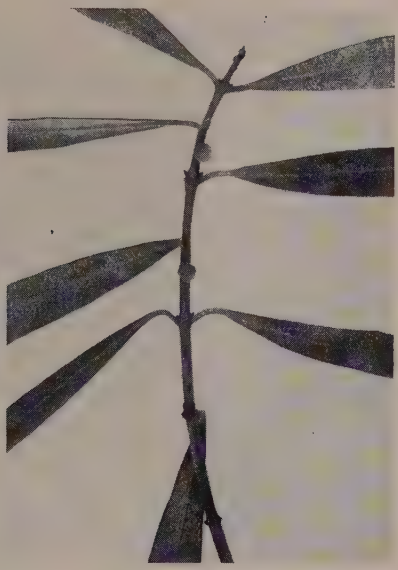

Fig. 4

Fig 3.—Galls on *Nerium oleander* leaf and stem caused by artifical inoculation with one culture used for cultural and physiological tests and morphology reported in text.

—Photo: H. E. Powell.

Fig. 4.—Galls on stem of *Olea europaea* caused by artifical inoculation with the same culture as on oleander in Fig. 3.

—Photo: J. W. Endt.

moriform, greenish-white galls formed slowly (Fig. 4). Check plants of oleander and olive inoculated with water did not form galls.

IDENTIFICATION OF ORGANISM

Identification of the causal organism was determined according to the morphology and the cultural and physiological reactions, suggested by the Society of American Bacteriologists (1946).

Morphology

The bacteria exist singly or in pairs as short rods with rounded ends. Single rods vary from 1.5μ to 2.1μ by 0.3μ to 0.4μ when stained with safranin. Using carbol fuchsin the measurements recorded were 1.5μ to 2.1μ by 0.45μ to 0.60μ. The rods are gram negative (Hucker's Modification) and are motile, having one to several polar flagellae (Gray's stain).

Cultural and Physiological Characteristics

In the following tests Difco media were used where applicable. A minimum of three tubes of three pathogenic strains was used for each test and all tubes were incubated at 25°C.

Beef Peptone Agar Colonies: circular, smooth, entire, translucent, greyish-white.

Beef Peptone Agar Slope: moderate, filiform, glistening, greyish-white, butyrous growth, musty odour, no pigment formation.

Beef Peptone Broth in 7 Days: moderate clouding, no surface growth, no odour, no sediment.

Potato Slice in 7 Days: moderate, filiform, glistening, cream coloured, butyrous growth, no additional odour, potato unchanged.

Nutrient Gelatin Stab 12%: thin, spreading surface growth; no liquefaction in 29 days.

Plain Gelatin Stab 12%: thin white surface growth, villous disc just beneath the surface and moderate filiform growth in stab; no liquefaction in 29 days.

Purple Milk: alkaline reaction without coagulation, tendency to clear slowly from the surface (3 mm. in 29 days), grey granular precipitate.

Nitrates (Beef peptone agar plus 0.1% KNO_3): no nitrite production in 2, 5, 7 or 14 days.

Hydrogen sulphide (Beef peptone agar plus 0.2% sodium thiosulphate and lead acetate strips): no hydrogen sulphide in 28 days.

Starch (Beef peptone agar plus 0.2% soluble starch): each test consisted of 2 bacterial streaks on three plates of each culture. One streak was wiped off agar before flooding with Lugol's solution. No hydrolysis in 2 or 7 days from the edges or under bacterial streaks.

Oxygen requirements: strict aerobe, i.e. no growth in 10 days on nutrient agar slopes covered with liquid paraffin and surface growth only in "poured" nutrient agar tubes.

Synthetic carbohydrate media 1% (using brom thymol blue as indicator): Media, adjusted before sterilization to give a pH of approximately 7.0 after sterilization, were dispensed in 3 ml. quantities in 4 x $\frac{1}{2}$ in. test tubes. Positive acid production was recorded when the pH of all 3 tubes of each culture was 6.3 or less. Checks were retained for the full period of test: no gas formed in any medium. Acid in arabinose, xylose, glucose, galactose, fructose, mannose, sucrose, glycerol and mannitol in 28 days; and in lactose in 52 days. No acid in rhamnose, maltose, raffinose, melezitose, starch, inulin, dextrin, or salicin in 52 days.

Taxonomy and Nomenclature of Organism

In 1908, Erwin F. Smith named *Bacterium savastanoi* as the cause of olive tubercle. Ferraris (1926) named *Bacterium tonellianum* as the cause of galls on oleander. C. O. Smith (1928) successfully inoculated the "oleander bacteriosis" organism into oleander, olive, and other hosts related to the olive. C. O. Smith also noted that cultural and morphological characteristics of the olive and oleander organisms "are not sufficiently different to separate the two into different species" and suggested the name *Pseudomonas savastanoi* var. *nerii* for the oleander knot organism. Adam and Pugsley (1934) after a limited morphological and physiological study of the oleander organism, stated that "the causal organism of oleander canker in Victoria is determined to be *Phytomonas tonellianum* (Ferraris) n. comb." Pinckard (1935) successfully

inoculated oleander-gall bacteria into pink and white oleanders and Manzanillo olive. He found that, of all the gall-producing bacteria, oleander and olive-knot organisms showed the greatest similarity. He retained the name *Ps. savastanoi* var. *nerii*. Breed *et al.* (1948) and Elliot (1951) described *Ps. savastanoi* var. *nerii* as a synonym of *Ps. tonelliana*. Dowson (1949) retains the name *Ps. savastanoi* f. sp. *nerii*.

On a basis of pathogenicity, morphology, and cultural and physiological reactions, the New Zealand organism causing oleander knot is comparable with *Ps. savastanoi* (Erw. F. Smith) Stevens, *Ps. savastanoi* var. *nerii* (C. O. Smith) and *Ps. tonelliana* (Ferraris) Burkholder. Any differences are small and can be attributed to strain variation, testing technique, or interpretation of results. It has been shown by others (C. O. Smith 1928, Pinckard 1935) that little difference exists in culture between the organisms causing oleander and olive knots, and that the oleander bacterium is pathogenic on olive. In view of this, and in accordance with the International Bacteriological Code of Nomenclature (Buchanan *et al.* 1948) there seems no reason why the organism causing oleander knot should be other than *Pseudomonas savastanoi* (Erw. F. Smith) Stevens.

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EFFECT OF STEAM-STERILIZED SOIL ON GROWTH OF REPLANT APPLE TREES

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(*Received for publication, 11 July 1956*)

Summary

The poor growth of apple trees on Northern Spy stock in soil that had previously grown trees on the same stock has been reproduced in pot experiments. Steam sterilization of this soil permitted as much growth as in fresh soil. Trees on Malling XII stock made satisfactory growth in soil that had previously grown trees on Northern Spy, but there was an indication of improved growth following soil sterilization. There was no sign of parasitic attack on roots of trees in either unsterilized or sterilized soil.

Introduction

Orchard experience in New Zealand has shown that an apple tree on Northern Spy rootstock, planted in the site where a tree on the same stock had previously grown, will make poor growth. This is an important point, as a large proportion of our existing apples are on this stock. The ability of a tree on another stock, such as Malling XII (Woodhead, 1946), to make satisfactory growth under these conditions has provided a means of overcoming this problem in the field. The factors responsible for the repression of growth of the trees on Northern Spy are not known, but some knowledge of these would help in evaluation of potential apple rootstocks. Some information has now been gained in an experiment where growth of trees was measured in sterilized and unsterilized soil from an old apple orchard.

Materials and Methods

Soil was a clay loam obtained from the Oratia Research Orchard near Auckland. Thirty-year-old Gravenstein trees on Northern Spy rootstock (confirmed by growth from a root cutting) had just been removed as they were severely infected with flat limb. Soil obtained from within a 3-ft. circle on these former tree sites is hereafter called "A" soil. Soil that had never grown apples was collected from between trees in an adjacent block of plums—this was designated "B" soil. Half of each batch of soil was sterilized by steaming at 200°–210°F. for ten minutes. Drums were filled with about 3 cu. ft. of either sterilized or unsterilized A or B soil.

In the winter, maiden apple trees, var. Sturmer Pippin, on either Northern Spy or Malling XII stock, were planted in these drums. There were three replications for each rootstock/soil/sterilization combination. These trees were kept outside and watered occasionally as required. Weeds that appeared in the unsterilized soil treatments were removed periodically. Following commercial practice, each tree was cut back after planting and each winter thereafter was pruned as if being shaped into an orchard tree. After three seasons' growth the experiment was discontinued as it was considered that further growth might result in some trees becoming potbound.

Before pruning each winter, the new linear growth made in the preceding season was measured. Total growth for each tree was the sum of lengths of all new growth made in that season.

RESULTS

Growth made by the trees in each of three seasons is given in Table 1.

TABLE 1. TOTAL LINEAR GROWTH (cm.) OF STURMER APPLE TREES IN EACH OF THREE SEASONS ON TWO ROOTSTOCKS IN STERILIZED AND UNSTERILIZED SOIL (Each figure is average for three trees)

Source of soil	Soil Treatment	Rootstock	1st Season's Growth	2nd Season's Growth	3rd Season's Growth
A	Unsterilized	Northern Spy	129	151	137
		Malling XII	224	228	250
	Sterilized	Northern Spy	204	276	285
		Malling XII	252	443	312
B	Unsterilized	Northern Spy	234	301	228
		Malling XII	291	438	347
	Sterilized	Northern Spy	291	261	223
		Malling XII	278	354	265
Differences required for significance		5%	103	90	74
		1%	141	124	102

A = soil from immediate vicinity of sites of old trees on Northern Spy stock.
B = soil of same type from adjacent block which had never grown apple trees.

The following results are apparent from this table.

1. Growth of trees on Northern Spy in A soil was significantly (at 1%) greater in sterilized than unsterilized soil, in the second and third seasons. Sterilization made no difference to trees on this stock in B soil.
2. Growth of trees on Malling XII in A soil was significantly (at 1%) greater in sterilized than unsterilized soil, in the second season only. In B soil, growth was greater in unsterilized soil in all seasons though only significantly (at 5%) in the third season.
3. For trees on both stocks growth in sterilized A soil was not significantly different from growth in unsterilized B soil.
4. With no sterilization, B soil produced significantly greater growth than A soil in trees on Northern Spy in all three seasons and in trees on Malling XII in the second and third season. With sterilization there is no significant difference between growth in A and B soils of trees on either stock.

At the end of the third season, inspection* of the root systems revealed no evidence of fungous, arthropod, or nematode infection in trees grown in any of the soils.

* I am indebted to F. J. Newhook and K. P. Lamb of Plant Diseases Division, Department of Scientific and Industrial Research, for these inspections.

DISCUSSION

"Growth" of a tree is not something easily measured. In the present work a single criterion—total length of new growth—has been used in each season as an assesssment of growth. This measurement takes no account of number of new shoots or of shoot diameter. Nonetheless it would seem to give a reasonable indication of the growth made by a tree.

The differences in growth, observed in this experiment, between trees on Northern Spy and Malling XII in unsterilized A soil are in agreement with field observations on the respective failure and success of these stocks as replants in sites where trees on Northern Spy have previously grown.

Sterilization of A soil improved the growth of trees on Northern Spy so that they were as vigorous as trees growing in unsterilized B soil. This suggests that steam sterilization overcame the factors responsible for poor growth.

Although, in the field, growth of replants on Malling XII stock is considered satisfactory, the present work indicates that sterilization of A soil may improve the growth of trees on Malling XII. In this connection it is also of interest that trees on Malling XII grew better in unsterilized B soil than in unsterilized A soil. It would seem that the factors responsible for the poor growth of replants on Northern Spy also affect replants on Malling XII but to a lesser extent.

Sterilization of B soil had no effect on growth of trees on Northern Spy stock but with trees of Malling XII stock there was a definite suggestion that growth in sterilized soil was not as good as in unsterilized. This might indicate that the increase in growth obtained with sterilization of A soil may be a net increase, being the resultant of a favourable and an unfavourable effect, as indicated by the results of sterilizing B soil.

Failure of trees on Northern Spy as replants is not a unique problem in fruitgrowing. In North America there is a similar problem with peaches (Upshall and Ruhnke, 1935). Proebsting and Gilmore (1941) thought that exhaustion of nutrients or carry over of diseases could not be responsible and produced evidence that toxic substances from roots of previous trees could effect growth of young peaches—a result that could not be confirmed by Harris and Gilkeson (1947). Shannon and Christ (1954) increased replant peach growth by fumigation with DD and thought that the improvement might be due to elimination of nematodes. Savage and Cowart (1954) could not associate poor replant growth with accumulation of pesticides or presence of old roots. Prince *et al.* (1955) obtained improved growth with applications of a high-magnesium lime and also with steam sterilization and fumigation. Chapman (1953) considered it likely that build-up of organisms detrimental to roots was the cause of replanting trouble with citrus in California.

Neither accumulation of spray materials nor depletion of nutrients would seem to be the cause of the trouble with trees on Northern Spy stock as it is unlikely that steam sterilization would rectify either of these conditions. Either a residual toxicity from the preceding crop or a build up of harmful organisms seems more likely. The present work has not distinguished between these two possibilities. Harmful organisms would

have been killed by the sterilization but, at the same time, toxic substances might also have been destroyed. The lack of any indication of fungous, arthropod, or nematode infection in the experimental trees makes it unlikely that the observed growth inhibitions would have been due to direct parasitic attack. The possibility of a more remote action by harmful organisms or attack by free-living nematodes have by no means been eliminated.

In the evaluation of new apple rootstocks in New Zealand one factor which has to be considered is the ability of a stock to grow as a replant succeeding a tree on Northern Spy stock. A technique such as was used in the present work should provide a reasonably easy testing procedure for the screening of potential stocks for this character without the need for extensive field trials.

Acknowledgement

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RESEEDING PASTURES BY OVERSOWING AND OVERDRILLING

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Summary

Experiments to compare the germination and establishment of short-rotation ryegrass reintroduced into a dairy-farm pasture by six different methods involving broadcasting, cultivating, and over-drilling, and to compare the subsequent pasture yields, are described.

Positive introduction of the seed into the soil by means of a furrow-opener attached to the coulter-tips of a standard hoe-coulter drill gave the quickest germination, the disc-coulter drill being next in order.

Seed broadcast following heavy surface cultivation, which destroyed practically all the existing species, gave the highest total yield during the winter following sowing but the lowest yield in the late spring.

Of the methods investigated, both the coulter tip attached to the hoe-coulter seed-and-fertilizer drill and the disc-coulter seed-and-fertilizer drill gave satisfactory results for a minimum of work and should provide a satisfactory method for the re-introduction of short-rotation ryegrass into clover-dominant pastures, provided the grazing management both before and after drilling is correct and fertility is adequate.

Introduction

Because of the tendency for short-rotation ryegrass to die out during a hot summer, a renewal of this grass is desirable, and annual oversowing rates as low as 2 lb. per acre have been recommended (Nielsen, 1950). Following the work of Hyde at Grasslands Division (Corkill, 1954), it is now a widely accepted practice in the North Island to reintroduce this grass into a pasture by broadcasting the seed on the surface and harrowing it in, or by broadcasting after varying degrees of surface cultivation. Seed is applied at rates up to 20 lb. per acre but results are far from satisfactory. Even where results have been good, it is apparent that a big wastage of seed results from broadcasting.

Exploratory trials in 1954 with short-rotation ryegrass and white clovers showed that, when using a disc-coulter drill for over-drilling existing pasture, a marked reduction in seeding rate was possible, while still providing a satisfactory balance between grass and clover in the subsequent pasture. Previous trials by Blackmore (1952) had established the method of introducing clover into tussock grassland by using a hoe-coulter drill fitted with a special furrow-opening tip. This promised to be satisfactory for overdrilling or "sod-seeding" run-out pastures. Trials conducted at Mississippi State College (Dudley and Wise, 1953) and at Sydney University (Breakwell and Jenkins, 1953; Breakwell 1954) also indicated possible improvements in existing techniques.

Following a very dry summer and early autumn in 1954–55, it was evident that in many areas replacement of short-rotation ryegrass would be necessary. It was then decided to lay down this trial to determine the efficiency of the different methods of oversowing likely to be used.

The field selected had been sown down 2 years previously with a mixture of 20 lb. short-rotation ryegrass, 6 lb. red clover, and 2 lb. of white clover per acre. By February 1955, there appeared to be a complete kill of the short-rotation ryegrass although both red and white clover had persisted.

The soil is described by N.Z. Soil Bureau (1954) as a grey-brown sandy loam to 13 in. overlying medium sand. Fertility was medium to high.

Experimental Procedure

Following a severe grazing, the field was topped to remove tall weed growth (mainly thistle and wild turnip) and harrowed with chain and tripod harrows to spread dung and residue from fed-out chou moellier and silage.

A plot trial was sown on 9 March, 1955. Short-rotation ryegrass was drilled or broadcast at the rate of 6 lb. of viable seed per acre except in Treatment 7. Two cwt. of serpentine superphosphate per acre was drilled with the seed or broadcast on all plots, including the controls.

The treatments comprised:

1. *Control*—no seed, fertilizer broadcast.
2. *Broadcast*—seed and fertilizer broadcast over existing pasture.
3. *Light Cultivation*—the pasture was harrowed once with weighted spike-tine harrows, seed and fertilizer broadcast, and harrowed again with the same harrows.
4. *Heavy Cultivation*—pasture disced three times with tractor tandem discs, spike-tine harrowed, chain harrowed, seed and fertilizer broadcast, and chain harrowed.
5. *Blackmore Coulter-tips*—seed and fertilizer drilled into existing pasture using a hoe-coulter drill fitted with Blackmore grassland oversowing tips, and chain harrowed.
6. *Disc Drill in 7-in. rows*—seed and fertilizer drilled into existing pasture using a standard disc-coulter drill sowing in 7-in. rows and chain harrowed.
7. *Disc Drill in 5-in. rows*—same as No. 6, but with the coulters spaced at 5-in. centres. No adjustment was made to the seeding rate, the rate per acre being ⅖ higher than all others.

The plots were arranged in a randomized-block design with four replications. Each pot was 7 ft. wide (one drill width) and 150 yards long. Five days after sowing, six permanent quadrats, each of 1 sq. yd., were pegged out in each plot. Except in Treatment 1, all volunteer grass seedlings were plucked out in these areas, each of which had been selected to avoid excessively weedy patches.

Results

Eight days after sowing, germination had taken place in the drilled plots (Treatments 5, 6, and 7) and seedlings were showing above ground. No emergence was noted in the other plots.

Two weeks after sowing, emergence counts were made on all plots. In the drilled plots the seedlings had in many cases reached the double leaf stage, whereas in the remainder they were much less advanced.

Six weeks after sowing, a further count was taken to determine the number of established plants in each plot.

At both counts, sequential analyses were performed so that the number of quadrats for examination could be determined from the data themselves. Six quadrats per plot were required at the first count, two per plot at the second.

The mean counts, their S.E.s, and summaries of the analyses of variance are given in Table 1.

TABLE 1. NUMBERS OF GRASS SEEDLINGS EMERGED PER SQ. YD. 2 WEEKS AND 6 WEEKS AFTER SOWING

Treatment	Number at 2 weeks (24 quadrats)	Number at 6 weeks (8 quadrats)	Ratio (from common quadrats)
1. Control	21†	35†	
2. Broadcast	6	36	
3. Light cultivation	169	221	1.21
4. Heavy cultivation	194	234	1.13
5. Blackmore tips	221	228	0.95
6. Disc drill 7 in.	186	207	1.08
7. Disc drill 5 in.	196‡		
S.E.s (Treatments 3–6)	± 11	± 13	±0.06
Sig. diff. at 5%	33	—	0.18
F(3,9 d.f.)	15.02**	0.54	3.70*
Coeff. of variation	25.2%	16.1%	14.7%

† Counts in Treatment 1 represent reseeding from old pasture only; in all other plots, counts represent emergence of seed sown plus volunteer seedlings emergd since removal on 14.3.55.

‡ 5/7 of actual count for comparison with other treatments.

** Significant at the 1% level of probability.

* Significant at the 5% level of probability.

Although the count in Treatment 5 was significantly higher than the mean of Treatments 3, 4, 6, and 7 at 2 weeks, there were no real differences (excluding Treatments 1 and 2) between the treatment means at 6 weeks. Climatic conditions during the period were very favourable to germination.

An analysis of the ratio of the two counts (the same quadrats being used for both) justified this conclusion. This is included in Table 1. The ratios were greater than unity in three of the treatments representing the additional emergence (volunteer + planted seed) and lower than unity in the fourth representing the balance between volunteer growth, a lower (or nil) further emergence of planted seed and death of established seedlings.

Plant Counts and Weights in Weedy and Non-weedy Areas

In order to obtain some information on the behaviour of the weed population, and its effect on the ryegrass under different treatments, weed counts and weights were taken from representative areas.

Chou moellier (*Brassica oleracea*) from a weed-infested source had been fed out on the trial area, with the result that very well-defined bands of weed, chiefly fathen (*Chenopodium album*), with some red root (*Amaranthus retroflexus*), and wild turnip (*Brassica campestris*) appeared across the trial area.

On 4 May 1955, two 2 sq. ft. quadrats were sited in each plot (one at random in the ordinary plot area, and one at random within one of the weedy bands) to determine the influence of severe weed infestation on the results obtained. In each marked quadrat, all ryegrass and fathen plants were harvested at ground level. Plant numbers, yields, and weight per plant were recorded.

All replications of Treatments 4, 5, and 6 only were examined.

Plant Counts

The means and their S.E.s, together with the differences significant at the 5% level of probability, are given in Table 2.

TABLE 2. NUMBER OF PLANTS (per 2 sq. ft.) IN WEEDY AND NON-WEEDY AREAS (4.5.55)

Treatment	Ryegrass		Fathen	
	Not weedy	Weedy	Not weedy	Weedy
4. Heavy cultivation	59	54	10 ± 1	530
5. Blackmore tips	55	17	—	370
6. Disc drill 7 in.	44	30	1	310
S.E.s of means	± 9	± 5	Not analysed	± 84
Sig. diff. at 5%	—	16		—
F (2, 6 d.f.)	0.78	16.34**		1.81
Coeff. of variation	34%	28%		42%

Ryegrass. In the non-weedy areas there were no differences, but in the weedy areas the numbers were significantly depressed in both of the drill treatments, those in Treatment 4 being similar in both weedy and non-weedy areas.

Fathen. It was observed that cultivation destroyed the old weed population and allowed a new one to replace it. In the non-weedy areas there was an increase in weed count, but in the weedy areas the new count was not significantly different from the old. It will also be noticed that this new high population has not depressed the ryegrass counts.

Weight per plant

The means, etc. are recorded in Table 3.

TABLE 3. WEIGHT OF D.M. PER PLANT (mg) IN WEEDY AND NON-WEEDY AREAS (4.5.55)

Treatments	Ryegrass		Fathen	
	Not weedy	Weedy	Not weedy	Weedy
4. Heavy cultivation	303	104	108	42
5. Blackmore tips	159	34	—	148
6. Disc drill 7 in.	133	57	98	165
S.E.s of Means	± 17	± 14	± 36	± 23
Sig. diff. at 5%	57	35	not analysed	81
F (2, 6 d.f.)	30.56**	6.16*		8.11*
Coeff. of variation	16.7%	44%		39%

Ryegrass. In both weedy and non-weedy areas the weight per plant was significantly greater in Treatment 4 than in either of the drill treatments. There were no real differences in weight per plant for the drill treatments 5 and 6 in either weedy or non-weedy areas. The effect of competition on the size of the individual plants is seen by reading across the lines of the above table. The earlier conclusion should be recalled here; that competition from weeds also cut down ryegrass numbers in Treatments 5 and 6.

Fathen. The weight of the individual fathen plants in the weedy areas of Treatment 4 was significantly lower than in the other treatments. This was simply because a new population was involved. (The 530 plants in Treatment 4 averaged 42 mg. while the 370 in Treatment 5 averaged 148 mg). In the non-weedy areas there was no significant difference in average fathen weight, although the conclusion here is of little moment as the numbers involved were small.

Species Yields

The means are recorded in Table 4.

TABLE 4. SPECIES YIELDS (D.M. (g.) per 2 sq. ft.) FROM WEEDY AND NON-WEEDY AREAS (4.5.55)

Treatments	Ryegrass		Fathen	
	Not weedy	Weedy	Not weedy	Weedy
4. Heavy cultivation	16.5	5.6	1.17	20.9
5. Blackmore tips	9.3	0.8	—	46.3
6. Disc drill 7 in.	6.2	2.4	0.12	44.1
S.E.s of Means	± 1.7	± 0.8	± 0.42	± 4.6
Sig. diff. at 5%	5.9	3.2	not analysed	16.0
F (2, 6 d.f.)	6.59*	9.71*		9.25*
Coeff. of variation	32%	54%		25%

Ryegrass. In both weedy and non-weedy areas there was a highly significant difference between the treatment means in favour of heavy cultivation. This will be referred to again when the plot yields are discussed. The suppression of ryegrass yield by weeds can be seen in any row of the above table.

Fathen. The weed production was significantly lower in the heavy cultivation treatment, since cultivation destroyed the old population of weeds and allowed them to be replaced by a new population of fathen plants, which were individually smaller at the date of sampling.

Dry Weight per Ryegrass Plant 8 Weeks after Sowing (4.5.55)

In each plot a point was chosen at random and the first 10 ryegrass plants in that row harvested. For each plot this was repeated 10 times to give 100 plants per plot. Each set of 10 plants was dried and weighed as a unit. This was performed for all replications of Treatments 4, 5, and 6 only.

The analysis of variance of the weights is given in Table 5.

TABLE 5. ANALYSIS OF VARIANCE OF DRY WEIGHTS PER 10 PLANTS

Source of Variation	d.f.	Mean Sq.	F and Result
Blocks	3		
Treatments	2	1.332	3.44(*)
Heavy cultivation v Drilling	1	2.646	6.83 *
Residual per plot	6	0.387	
Samples within plots	108	0.681	

Coeff. of variation per plot of 100 plants = 10.2%
(*) significant at the 10% level of probability.

The treatment means, their S.E.s etc. are as follows:

4. Heavy cultivation	2.15 ± 0.10 g.	Sig diff. at 5% = 0.34
5. Blackmore tips	1.82 „	
6. Disc drill 7 in.	1.85 „	

The means for Treatments 5 and 6 were both significantly lower than that for Treatment 4 at the 8% level of probability, the test for Heavy cultivation v. Drilling in the analysis of variance showing that the advantage of Heavy cultivation was significant at the 5% level.

Thus, at the beginning of May, the individual plants were significantly bigger in the Heavy-cultivation treatment than in the others. This must be interpreted in the light of counts of ryegrass, weeds, and clovers, and their relative development, data on which have been provided.

The last line in the analysis of variance was for the purpose of determining the number of samples required had the 100 taken per plot been insufficient, or to determine whether any further sampling would be worthwhile. It shows that the variation within plots was serious, but that the overall variation between plots was relatively small. This

was not unexpected, as it was observed that the plots consisted of a large number of small patches of weed in a mosaic.

Species Yields and Botanical Composition

The establishment trial was followed by a series of herbage-yield comparisons so that the behaviour of the different treatments could be determined.

Cuts (four in all) were taken each time the growth on the plots reached a stage suitable for grazing with cattle. No variation in the grazing dates for different treatments was possible, the area being considered suitable for cutting and grazing when the grass in the majority of the plots had reached 6 in. to 9 in. in height.

The common grazing dates used meant that this part of the trial could not be carried on indefinitely without becoming invalid, since grazing intervals would not be optimal for all treatments. The errors introduced by fertility transfer at each grazing also limit the accuracy and useful duration of this part of the trial, but such transfer effects would be expected to reduce true treatment effects, results remaining conservative even if affected.

After each cut (a different location being used on each occasion) the whole trial area was grazed quickly with cattle, then topped with a mower and chain harrowed to spread dung.

On each occasion an area 40 ft. long was cut with a 5 ft. mower, thus giving 200 sq. ft. from each plot. The green material was weighed, and samples taken from each plot for dry-matter determinations and botanical analysis. The species and total yields, botanical compositions, and summaries of the analyses of variance are recorded for the 4 cuts in Tables 6–9 respectively, and graphically in Figures 1–3.

First Cut, 10 May

TABLE 6. Species Yields (lb. D.M. per acre) and Composition at First Cut 10.5.55

Treatments	Species Yields				Percentages		
	Ryegrass	Clover	Other Species	Total	Ryegrass	Clover	Other Species
1. Control	120	390	455	965	12.5	41.0	46.5
2. Broadcast	85	440	360	885	10.0	52.0	38.0
3. Light cultivation	460	195	350	1005	47.5	19.0	33.5
4. Heavy cultivation	655	50	535	1240	52.5	4.0	43.5
5. Blackmore tips	435	380	510	1325	33.0	29.0	38.0
6. Disc drill 7 in.	390	330	345	1065	36.0	32.0	32.0
7. Disc drill 5 in.	510†	345†	385†	1240†	39.5	29.5	31.0
S.E.s of means	±78	±41	±74	±78	±6.3	±3.4	±5.8
Sig. diff. at 5%	233	122	—	233	18.7	10.0	—
F (6, 18 d.f.)	46.9**	10.9**	1.13	4.44**	6.66**	21.10**	1.01
Coeff. of variation	42%	27%	35%	14%	38%	23%	31%

† Effective seeding rate was 40% higher in Treatment 7. No adjustments have been made in this or following tables.

Ryegrass. The failure of the broadcast treatment is seen in both yields and percentages where the results were significantly lower than for all other treatments and no better than Control. Of the remaining treatments the Heavy cultivation was significantly higher than the Blackmore or Disc drill in yield and in ryegrass percentage (a function of the clover percentage—see below).

Clover. It is clear that the effect of cultivation has been to reduce clover yields. Heavy cultivation gave the greater reduction, Light cultivation being less severe. None of the remaining clover yields differed significantly among themselves or from Control. That is, none of the drill treatments have affected the yield of clovers at the first cut. The clover percentages recorded are clearly determined by increasing reduction of clover content by cultivation and the lack of ryegrass in Treatments 1 and 2.

Other Species. There were no differences in yield at this stage. It has already been shown that in the very weedy patches heavy cultivation replaced a heavy infestation of old fathen plants by an equally heavy (or heavier) infestation of new and smaller fathen plants, and that in the less weedy areas of the same plots the small population of fathen plants was significantly increased by cultivation. When the plots were cut and weighed (one week later than the weed-patch examination) there were no significant differences in weed yield, weed here including fathen together with wild turnip and chickweed (*Stellaria media*). The difference between Light cultivation and Heavy cultivation was almost significant at this stage, and may perhaps be due to the greater reduction of competition from clovers in the Heavy cultivation, and/or the greater turn-up of buried weed seed in that treatment. There were no significant differences in weed percentage at this stage.

Total Production. The Blackmore tips and Heavy cultivation did not differ significantly, but both were significantly higher than either the Light cultivation or the Disc drill. The Light cultivation was not significantly better than Control at this stage (or the Broadcast, which was virtually a second Control) for the increase of ryegrass was mainly balanced by the loss in clover. Very little can be said about Treatment 7 at this cut, owing to the different seeding rate. The yields of clover and other species cannot be compared since the effects of inter-species competition cannot be determined. It can be said that, whatever the causes, the yield of ryegrass from the heavier sowing was not significantly greater than the average of the other drilled and cultivated sowings. In total yield, Treatment 7 was not significantly different from Treatments 4 and 5, but was almost significantly higher than Treatment 6—Disc drill at 7 in.

Second Cut, 10 August

Ryegrass. The means of yields fell into four groups, each significantly different from the others; (a) Heavy cultivation, (b) Light cultivation, and 5 in. Disc drill, (c) 7 in. Disc drill and Blackmore tips, (d) Control and Broadcast. In ryegrass percentage, the Control and Broadcast means were significantly lower than all others, but there were no other significant differences.

Clover. In yield, the differences did not reach significance level, as the amount of clover was low in all plots and variation within treat-

TABLE 7. SPECIES YIELDS (lb. D.M. per acre) AND COMPOSITION AT SECOND CUT 10.8.55

Treatment	Species Yields			Percentages	
	Ryegrass	Clover	Total	Ryegrass	Clover
1. Control	155	10.5	205	70.5	5.1
2. Broadcast	90	10.0	120	78.5	8.1
3. Light cultivation	790	12.5	835	94.0	1.0
4. Heavy cultivation	1330	8.0	1390	95.5	0.6
5. Blackmore tips	570	17.0	615	93.5	2.6
6. Disc drill 7 in.	465	22.5	515	90.0	4.5
7. Disc drill 5 in.	740	12.5	805	91.0	1.5
S.E.s of means	± 92	± 4.1	± 95	± 2.8	± 0.9
Sig. diff. at 5%	275	—	281	†	†
F (6, 18 d.f.)	20.7**	1.45	20.5**	9.39**	6.95**
Coeff. of variation	31%	61%	30%	7.5%	30%

† Analyses were performed and conclusions drawn using transformed data; average S.E.s are given only.

ments high (coefficient of variation 61%). In clover percentage the means were generally in reverse order compared with ryegrass; that for the Broadcast treatment was significantly highest, and those for the two cultivation treatments significantly lowest. Although there were no significant differences between the clover yields, there were interesting differences in the ryegrass: clover ratios. These were as follows:

1. Control	15 : 1
2. Broadcast	10 : 1
3. Light cultivation	76 : 1
4. Heavy cultivation	239 : 1
5. Blackmore tips	43 : 1
6. Disc drill 7 in.	38 : 1
7. Disc drill 5 in.	73 : 1

Other Species. The yields and percentages of other species were recorded, but as there were no significant differences, owing to the patchiness of the weed, these are not given in this summary.

Total Production. As was to be expected from the lack of significance in the clover means, and their low level throughout, the conclusions in total production are exactly the same as those for ryegrass, and will not be repeated.

Third Cut, 19 September

Ryegrass. In yield, Control was clearly lower than any other treatment, but had increased markedly in comparison with previous cuts. The Broadcast mean was significantly lower than the averge of the remaining sown treatments, but did not differ significantly from the Heavy-cultivation treatment. The remaining 4 treatments did not differ significantly. Of the sown treatments, Heavy cultivation gave the highest yield of ryegrass in the August cut, but the lowest in the September cut. In percentage, the treatments fell into 4 significantly different groups

TABLE 8. SPECIES YIELDS (lb. D.M. per acre) AND COMPOSITION AT THIRD CUT 19.9.55

Treatment	Species Yields				Percentages	
	Ryegrass	Clover	Other Species	Total	Ryegrass	Clover
1. Control	387	246	123	756	51.0	32.5
2. Broadcast	515	212	153	880	58.0	24.0
3. Light cultivation	650	102	55	807	80.5	12.5
4. Heavy cultivation	510	16	33	559	90.5	3.5
5. Blackmore tips	718	182	50	950	75.0	19.5
6. Disc drill 7 in.	753	156	46	955	79.0	14.5
7. Disc drill 5 in.	712	134	62	908	78.0	15.0
S.E.s of means	± 55	± 25	± 20	± 52	± 3.4	± 2.5
Sig. diff. at 5%	163	76	59	156	†	†
F (6, 18 d.f.)	6.30**	8.95**	5.19**	7.20**	16.49**	15.18**
Coeff. of variation	18%	34%	53%	12.6%	7.5%	17%

† Analyses were performed and conclusions drawn using transformed data; average S.E.s are given only.

(a) Heavy cultivation, (b) all other Drilled and Cultivated treatments, (c) Broadcast, (d) Control.

Clover. Here the means fell into three distinct groups; (a) Treatment 4 was significantly lowest in yield and percentage, (b) Control and Broadcast were significantly the highest, with Control significantly the higher in percentage, (c) of the remaining treatments Light cultivation was significantly lower than the Blackmore tips both in yield and percentage.

Other Species. Here there were two distinct groups. Control and Broadacst were both significantly higher than all others, among which there were no real differences.

Total Production. The Heavy cultivation was distinctly the lowest at this cut. The Control and Light cultivation did not differ significantly but both were significantly lower than the overall mean of the three drill treatments. The remaining four treatments did not differ significantly, obviously because the higher weed content balanced a significantly lower ryegrass content.

Comparison of the yields in August and September shows: (a) increased yields of both ryegrass and clover in the Control and Broadcast treatments, (b) a slump in ryegrass in the plots with little clover, so that the best treatments in August were the worst by September, and (c) increasing weed in the plots with poor ryegrass.

Further, at the September cut the three drilled treatments did not differ significantly in either ryegrass or total production, the seeding-rate difference for Treatment 7 having disappeared from all the data for this treatment.

Considerable changes in clover percentage occurred between the August and September cuts. In August all clover percentages were low, with the Broadcast highest at 8%, and the cultivation treatments

significantly lowest at 1%. By September these differences had become more marked as the clover percentages had increased in all plots, the Heavy cultivation clover rising to 3% only, while that in the Control rose to 32%, with the remaining treatments in an intermediate position of 15–20% clover.

Fourth Cut, 10 November

TABLE 9. SPECIES YIELDS (lb. D.M. per acre) AND COMPOSITION AT FOURTH CUT 10.11.55

Treatment	Species Yields				Percentages	
	Ryegrass	Clover	Other Species	Total	Ryegrass	Clover
1. Control	2050	655	280	2985	68.0	22.0
2. Broadcast	2590	360	195	3145	82.5	11.5
3. Light cultivation	2860	315	90	3265	87.5	9.5
4. Heavy cultivation	2180	330	110	2620	83.0	12.5
5. Blackmore tips	2760	570	135	3465	79.5	16.5
6. Disc drill 7 in.	2500	395	125	3020	81.5	15.0
7. Disc drill 5 in.	2960	300	185	3445	86.0	9.0
S.E.s of means	± 209	± 72	± 60	± 171	± 3.9	± 2.8
Sig. diff. at 5%	622	216	—	510	11.6	8.5
F (6, 18 d.f.)	2.40(*)	3.70*	1.17	2.95*	2.73*	2.68*
Coeff. of variation	17%	35%	74%	11%	10%	42%

Ryegrass. Control and Heavy cultivation were significantly low, but there were no other real differences. In ryegrass percentages, Control was significantly lower than all other treatments.

Clover. In clover yield the Control was higher than all others with the exception of the Blackmore treatment, and the latter was significantly higher than either Cultivation treatment. In percentage, the Control mean was significantly highest.

Other Species. Control mean was significantly higher than the overall mean of the treatments, but there were no other real differences, partly because of the very high variability.

Total Production. The mean for Heavy cultivation was still significantly lower than the means for the best treatments. Because of the large weed and clover components in Control, this treatment, if worse than any other, was worse than only the best treatments.

Clover percentages dropped in all treatments except Heavy cultivation following the September cut. Heavy-cultivation clover percentage rose from 3.5% to 12.5%.

The changes in ryegrass percentages were the reverse of those of the clovers. There was a rise in ryegrass percentage in all treatments except Heavy cultivation where there was a slight drop. The rise in ryegrass was most marked in Control (from 51% to 68%) and Broadcast (58% to 82%), these having previously the two lowest percentages and Heavy cultivation the highest. There appears therefore to have been a levelling out of percentages for both ryegrass and clover.

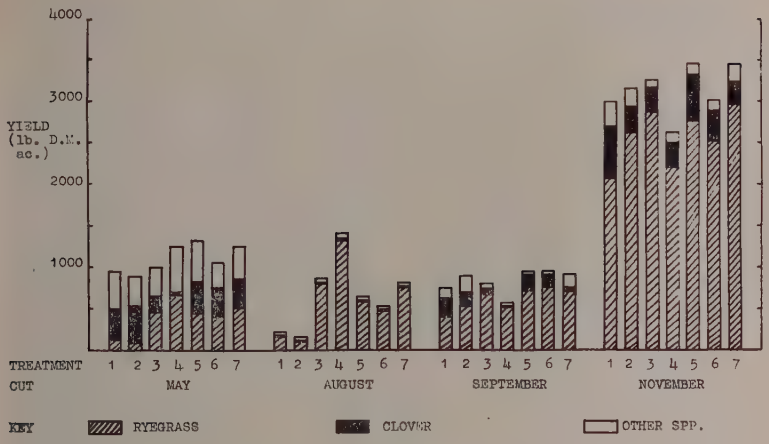
4000
3000
YIELD (lb. D.M. ac.)
2000
1000
TREATMENT 1 2 3 4 5 6 7 1 2 3 4 5 6 7 1 2 3 4 5 6 7 1 2 3 4 5 6 7
CUT MAY AUGUST SEPTEMBER NOVEMBER
KEY RYEGRASS CLOVER OTHER SPP.

Fig. 1.—Individual yields (lb. D.M. per acre) for all treatments.

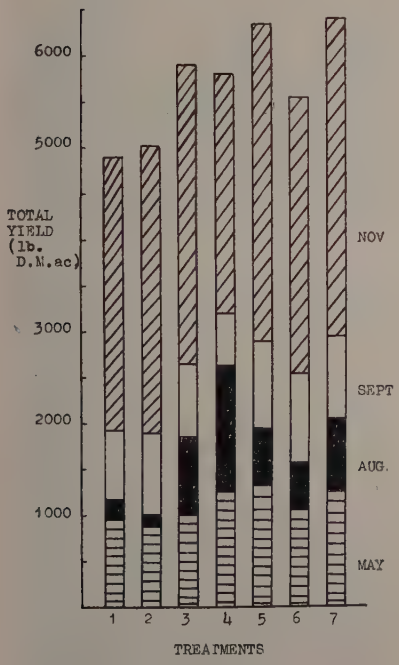

Fig. 2.

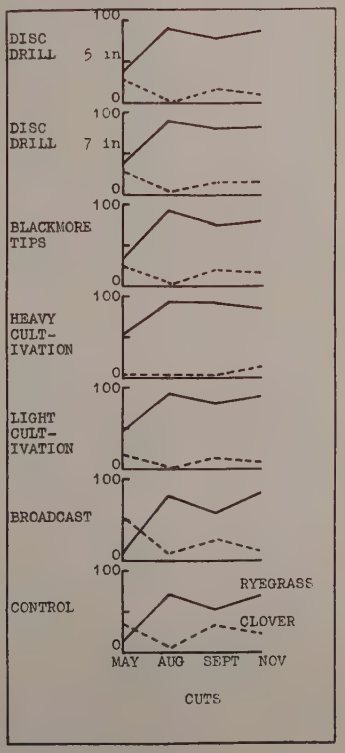

Fig. 3.

Fig. 2.—Total yields (lb. D.M. per acre) for all treatments 9 March to 10 November 1955. (Seed rate per acre in Treatment 7 (Disc Drill 5 in.) was 40% higher than in the others.)

Fig. 3.—Fluctuations of clover and ryegrass, as percentage of the total yield.

Fig. 4.—Area prior to overdrilling, 22 February 1955. *Photo: H. Drake.*

Fig. 5.—Typical result from overdrilling: Treatment 7 (Disc drill at 5 in.) and Control. 18 May 1955. *Photo: H. Drake.*

Conclusions and Recommendations

1. In clover-dominant pastures, where fertility is at a reasonably high level, to provide maximum yield and a well-balanced sward of short-rotation ryegrass and clover in the spring following overdrilling of the grass, it is necessary to reduce competition from the established species and to provide optimum germinating and growing conditions for the introduced ryegrass in the early stages. This may be achieved by a hard grazing, followed by a light cultivation to reduce clover percentage as in Treatment 3, and drilling of the seed and fertilizer as in Treatments 5 and 6. Subsequent grazing management should differ from normal new-pasture management until the following spring. Lax grazing will allow the vigorous short-rotation ryegrass to become thoroughly established and well tillered.

2. If clover dominance is not marked and fertility is adequate, successful results will be obtained by hard grazing immediately before overdrilling with seed and fertilizer, and continuing until germination. Grazing management that favours the grass should again be followed during the early stages.

3. If overdrilling is practised early in the autumn and under good fertility conditions, a seeding rate of 6 lb. per acre of viable short-rotation ryegrass will provide sufficient plants to ensure grass dominance. If good seedbed conditions are provided by light or heavy cultivation similar results can be expected, but climatic conditions favourable to germination are necessary if the seed is broadcast rather than drilled. Broadcasting without any cultivation can produce very indifferent results.

4. The poorer results from the Disc-drill treatments relative to Blackmore tips are attributed to the fact that the slice of soil removed by the disc blades frequently fell back into its original position and that there was no positive control of sowing depth. The plants were trapped under this slice and had difficuty in emerging. Even when they did emerge, after underground growth in many cases up to 6 in., the plants were so exhausted that many failed to survive the competition from established species. Where the disc slice had been completely removed and the seed covered by a light layer of soil (provided by the chain harrows) emergence was as rapid and even as with Treatment 5 (Blackmore tips).

5. Areas heavily infested with weed require sufficient surface cultivation to destroy existing weeds, and the grass-seed may then be drilled, or broadcast if conditions are favourable. New weed seedlings will offer less competition than will established weed during the early stages of growth and could be checked by mowing (or spraying) after the first grazing.

6. If the field has been heavily surface cultivated it would be necessary to drill (or broadcast if conditions are favourable) both grass-seed and clover in the autumn to provide the clover necessary for vigour of the pasture in the spring (see Treatment 4, Tables 7 and 8).

7. The satisfactory establishment, together with the minimum of work, makes overdrilling with a machine which places the seed and fertilizer together in the soil in a shallow open groove a most attractive and practicable method.

(A later modification to the disc drill has been the addition of a simple depth-skid to each disc coulter. This skid limits penetration of the disc to the desired depth, irrespective of increased spring pressure, and so allows a furrow of constant depth to be cut through varying types of surface cover or soil. If the soil is reasonably dry and a forward speed of about 6 m.p.h. is maintained with the depth-skid fitted, the slice of soil will be completely removed under most conditions, and trailing chain harrows will provide sufficient cover for the seed.)

Acknowledgements

The authors wish to acknowledge helpful discussions with Mr. E. O. C. Hyde and Dr. P. D. Sears of Grasslands Division, Department of Scientific and Industrial Research, and the technical assistance given by that Division, particularly by the Herbage Section under Miss D. P. Owen. Grateful acknowledgement is made to Mr. S. L. Green, Manager of the Massey College Dairy Farm, for his co-operation in the grazing management during the trial period.

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THRIPS INFESTING COCKSFOOT IN NEW ZEALAND

III. THE IDENTITY OF TWO SPECIES OF *Chirothrips*.

By K. M. Doull, Waite Agricultural Research Institute, University of Adelaide

(*Received for publication, 29 August 1956*)

Summary

Further studies have shown that two species of *Chirothrips* occur on cocksfoot in New Zealand. *Chirothrips manicatus* Haliday, previously given the common name of cocksfoot thrips is now found to breed mainly on meadow foxtail, and is comparatively rare on cocksfoot. The common species on cocksfoot is *Chirothrips pallidicornis* Priesner. Characters which serve to separate the two species are described and figured.

Further studies of *Chirothrips* collected from grasses in New Zealand have been made by Mr. E. R. Speyer at the British Museum and by the author. Two species may be distinguished.

Chirothrips manicatus Haliday.

This species which was previously given the common name of "Cocksfoot Thrips" (Doull 1956) occurs only occasionally on this plant, its main host at Lincoln being meadow foxtail. *C. manicatus* is a particularly variable species. None of the characteristics based on differences in length of postero-angular setae of the prothorax, the arrangement of circular tubercles on the posterior margins of abdominal sternites, or on chaetotaxy of the hind vein of the forewing can be used with certainty to separate it from other species of the same genus.

Adults of *C. manicatus* may be distinguished by the following characters:

Females:

Antennal segments II to V as dark in colour as segment I.

Outer margin of antennal segment II concave in outline in centre and slightly convex towards apical projection.

Projection of head in front of each eye at least 10μ long.

Hind vein of forewing with three or four, sometimes as many as six setae.

Males:

Circular glandular depressions on abdominal sternites 3 to 7.

Chirothrips pallidicornis Priesner.

This species was first described by Priesner (1928) as *C. manicatus* forma *pallidicornis*. In 1938 the same author described it as a new species. It dominates the thrips collected from cocksfoot but occurs only in very small numbers in material from meadow foxtail. Adults of *C. pallidicornis* may be distinguished by the following characters:

Females:

Antennal segments II to V much lighter in colour than segment I.

Outer margin of antennal segment II straight in outline.

Projection of head in front of each eye not more than 5μ long.

Hind vein of forewing *usually* with five setae.

Males:

Circular glandular depressions on abdominal sternites 3 to 6.

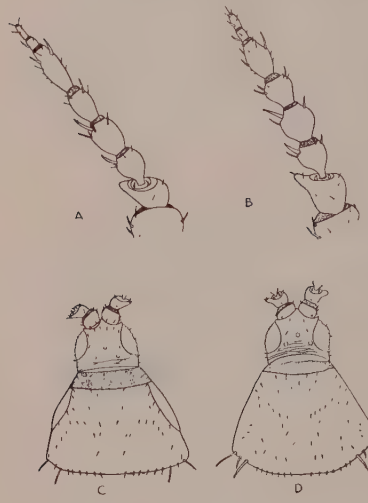

Fig. 1

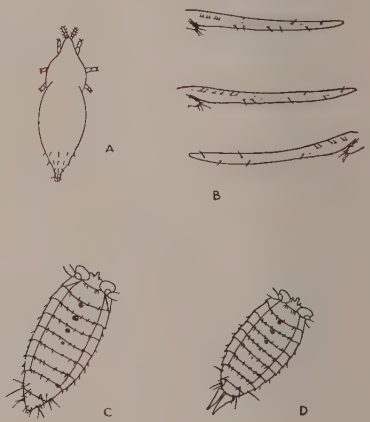

Fig 2

Fig. 1—A. Left antenna *C. pallidicornis* female. B. left antenna *C. manicatus* female. C. Head and prothorax *C. pallidicornis* female. D. Head and prothorax *C. manicatus* female.

Fig. 2.—A. Larva II *C. pallidicornis*. B. Right forewing *C. pallidicornis* (Upper); *C. manicatus* (Middle); left forewing *C. manicatus* (Lower). C. Abdominal sternites *C. pallidicornis* male. D. Abdominal sternites *C. manicatus* male.

DISCUSSION

The characters set out above refer only to adults of *manicatus* and *pallidicornis* and may not serve to separate them from other species of the genus. No characters have been found which serve to separate immature stages of the two species.

It is of interest to note that *C. pallidicornis* in N.Z. shows a high degree of adaptation to cocksfoot. At Lincoln where cocksfoot and meadow foxtail grow together in the same field, the number of *pallidicornis* found on meadow foxtail is so small as to suggest that they are only accidental visitors to that plant. Similarly *manicatus* is quite rare on cocksfoot.

Until it becomes possible to separate immature stages of the two species there will be no way of telling whether *C. pallidicornis* does in fact breed on foxtail or *C. manicatus* on cocksfoot. There is no apparent reason why this should not be so. But foxtail flowers earlier than cocksfoot and this specificity may in fact be due to earlier emergence of hibernating females of *manicatus* or perhaps to earlier maturation of their ovaries.

Details of the biology of *C. manicatus* as described in the previous paper should now be taken to apply to *C. pallidicornis*. Field observations indicate that the biology of *manicatus* on meadow foxtail is similar to that of *pallidicornis* on cocksfoot.

ACKNOWLEDGEMENTS

Acknowledgements are gratefully made to Mr. E. R. Speyer for time and trouble taken in identifying the two species; to Mr. D. C. Swan for helpful criticism of the manuscript and to Miss R. Dickinson for the illustrations.

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NEW RECORDS OF FUNGOUS DISEASES IN NEW ZEALAND 1955-56

By Joan M. Dingley, and R. M. Brien, Plant Diseases Division, Department of Scientific and Industrial Research, Auckland

(Received for publication, 23 July 1956)

Summary

Sorosphaera veronicae Schroeter on *Veronica tournefortii* Gmel., *Sphaerotheca humuli* var. *fuliginea* Schlecht on *Coriaria angustissima* Hook. f. and on *C. thymifolia* Humb. et Bonpl., and *Puccinia iridis* (DC.) Wallr. on *Iris xiphium* (Dutch iris) are recorded for the first time in New Zealand.

Additional host records include *Peronospora grisea* (Unger) de Bary on *Hebe hulkeana* (F. Muell. ex Hook) Ckn. et Allan, *Elsinoe piri* (Woron). on *Pyrus communis* L. (pear) *Erysiphe polygoni* (DC.) emend. Salmon on *Aristotelia fruticosa* Hook. f., *Oidium* sp. on *Dahlia* sp. cult., *Physalospora obtusa* (Schw.) Cke. on *Prunus persica* L. (peach) and on *Rubus idaeus* L. (raspberry), *Uromyces appendiculatus* (Pers.) Link on *Phaseolus coccineus* L. (scarlet runner bean), and *Stagonospora curtisii* (Berk.) Sacc. on *Hippeastrum* sp. and *Nerine* spp.

The following notes are based on additional records made during the last twelve months of material collected throughout New Zealand. In all cases specimens have been placed in the herbarium of the Plant Diseases Division, Auckland, New Zealand.

1. *Peronospora grisea* Unger, Bot. Ztg. *5;* 315; 1847.
On *Hebe hulkeana* (F. Mueller ex Hook.) Ckn. et Allan, Mt. Albert, Auckland, J.M.D., Feb. 1955.

Conidiophores densely aggregated on under surfaces of leaves, 200-250 × 5.5-7μ, dichotomously branched 5-6 times, ending in curved prongs. Conidia globose or broadly oval, 14-15 × 14-15.5μ; oospores not present in New Zealand collections.

The fungus discoloured and distorted leaves and young shoots of the *Hebe*. This species has been recorded in New Zealand on *Veronica arvensis* (field speedwell) by Brien and Dingley (1955). Gaumann (1923), separated this species on speedwell as *P. verna* Gaumann; it is interesting to note that infected plants of speedwell were found growing among the *Hebe* the season before it became infected. Massee (1914) recorded *P. grisea* on *Veronica hulkeana* F. Mueller ex Hook, (*Hebe hulkeana*) in Great Britain.

2. *Sorosphaera veronicae* Schroeter, J. in Cohn, F.J. "Kryptogamen Flora von Schlesien" Vol. 3, p. 135; 1886.
On *Veronica tournefortii* Gmel., Mangere, Auckland, P. Fry, Aug. 1955 (Fig. 1)..

Galls 0.5-2.0 × 0.2-0.5 cm. forming series of swellings on stems, petioles, and leaves. Cytosori compacted in swollen parenchyma cells among poorly developed vascular bundles. Cytosori light brown, globose, or oval, in hollow spheres, 12-30 × 12-40μ, 25-30 compacted in a single parenchyma cell of the host. Cytosori formed of 16-64 resting spores, 4-5.5 × 4-6μ. Resting spores germinate to form bilflagellate zoospores.

Fig. 1.—Galls on *Veronica tournefortii* caused by *Sorosphaera veronicae*. —*Photo: J. W. Endt.*

The first collection was found growing in a market garden at Mangere; examination of other speedwell plants growing in the vicinity of Auckland showed that this gall-forming organism was not uncommon. Karling (1942) recorded it from Europe, Great Britain, and North America.

3. *Elsinoe piri* (Woronich.) Jenkins, J. agric. Res. *44:* 696; 1932.
On *Pyrus communis* L., Henderson, Auckland, A. J. Hiam, Feb. 1955 (Fig. 2).

Elsinoe piri was recorded on apples (*Pyrus malus* L.) by Brien and Dingley (1951); pear leaves showing similar symptoms were collected from Henderson. The disease lesions formed only conidial fructifications.

4. *Erysiphe polygoni* DC, emend. Salmon, Mem. Torrey bot. Cl. *9:* 175; 1900.
On *Aristotelia fruticosa* Hook. f., Arthur Pass, Canterbury, J.M.D., Jan. 1956.

Fig. 2.—*Elsinoe piri* on pear leaves.
—*Photo: J. W. Endt.*

Perithecia superficial or semi-immersed in a mycelial mat, globose or oval, dark brown when mature, 70-120μ diameter, with few unbranched appendages; 2-4 asci present in each perithecium. Asci clavate, shortly stalked, rarely sessile, 40-90 × 25-40μ 6-8 spores irregularly arranged in ascus; spores oval, 16-20 × 9-10μ, hyaline. Perithecia were present in the mycelial mat on both leaves and fruit. Berkeley (1855) described *E. densa* from specimens of powdery mildew on *Aristotelia racemosa* Hook. f. (syn. *A. serrata* (Forst.) Oliver). Salmon (1900) examined Berkeley's material in the Kew herbarium and found them to be identical with *E. polygoni*. This fungus, collected from Canterbury on *Aristotelia fruticosa*, agrees with Salmon's description.

Erysiphe polygoni is common on a wide range of host plants but perithecia are rarely found.

5. *Sphaerotheca humuli* var. *fuliginea* (Schlecht) Salmon, Mem. Torrey bot. Cl. *9:* 49; 1900.
On *Coriaria angustissima* Hook. f. and *C. thymifolia* Humb. et Bonpl., Arthur Pass, Canterbury, J.M.D., Jan. 1956.

Perithecia more or less superficial on a mycelial mat, globose or oval, dark brown, 75-85μ diameter, with long, septate, unbranched appendages; perithecial wall consisting of large irregular cells, 25-30 × 20μ; perithecia containing a single ascus. Asci more or less globose, sometimes oval, 55-75 × 50-60μ, 8-spored, spores oval or globose, 15-18 × 12-16μ.

Perithecia were present on both leaves and fruit. The large irregular cells of the outer perithecial wall were used by Salmon to separate this variety; they are characteristic of perithecia in this collection.

6. *Oidium* sp.

On *Dahlia* sp. cult., Remuera, Auckland, J.M.D., March 1956.

During autumn 1956 the *Oidium* stage of a powdery mildew was common on cultivated species of *Dahlia* throughout the Auckland district.

7. *Physalospora obtusa* (Schw.) Cooke, Grevillea *20:* 86; 1892.
On *Prunus persica* L., Whangarei, Auckland, P. Everett, Oct. 1948.
On *Rubus idaeus* L., Motueka, Nelson, P. J. Brook, June 1955.

This fungus was first recorded in New Zealand by T. Kirk (1907) on apple and later by Cunningham (1921) on pear and quince (*Cydonia oblonga* Mill.). In the above collections the fungus is associated with die back of young shoots. Further collections were made from peach in the Auckland district this season (1956).

8. *Uromyces appendiculatus* (Pers.) Link, Mag. Ges. naturf. Freunde Berlin, *7:* 28; 1816.
On *Phaseolus coccineus* L., Owairaka, Auckland, J.M.D., March 1955.

French-bean rust was recorded by Brien and Jacks (1954) on varieties of *Phaseolus vulgaris* L., e.g. Sydney Wonder, Fardenlosa; they recorded that, while artificially inoculated varieties of *P. coccineus* (scarlet runner) were susceptible, no record of natural infection was obtained. In the Auckland district in autumn 1955, and again this year, scarlet-runner beans have been heavily infected with rust.

9. *Puccinia iridis* (DC.) Wallr. in Rabenhorst, G. L. "Deutschlands Kryptogamen-Flora" (1st ed.) Vol. 1, p. 23; 1844.
On *Iris xiphium* L., Palmerston North, Wellington, C. L. Napier, March 1956.

O.I. unknown.

II. Uredosori mostly hypophyllous, numerous, scattered, sometimes confluent, then up to 1mm. long; uredosori 0.25-0.5mm. rupturing by a longitudinal slit in epidermis, cinnamon, surrounded by the ruptured epidermis. Spores sub-globose or oval, 19-28 × 17.5-24μ, epispore 2-3μ thick, closely echinulate, cinnamon, 2-4 germ pores usually equatorial.

III. Teleutosori hypophyllous, scattered, sometimes confluent, up to 2 mm. long, sori naked, rupturing through a longitudinal slit. Teleutospores clavate or oblong, rounded, rarely apiculate, but thickened at the apex, often constricted 38-55 × 10-21μ, epispore smooth, 2-4μ thick but at the apex up to 15μ thick. Pedicels persistent, stout, pigmented 15-25 × 5-7μ.

The species is recorded from Europe and North America. Infected plants were grown from bulbs recently imported from Holland. It is recorded in North Amercia and Europe as attacking a wide range of species of *Iris*.

10. *Stagonospora curtisii* (Berk.) Sacc., Syll. Fung. *3:* 451; 1884.
On *Hippeastrum* sp., Onehunga, Auckland, A. A. Coates, Dec. 1955.

On *Nerine curvifolia* var. *fothergillii* Baker, Remuera, Auckland, J.M.D., June 1956.
On *Nerine* sp., Palmerston North, Wellington, C. L. Napier, June 1956.

Stagonospora curtisii was recorded from New Zealand on *Narcissus* sp., by Brien (1942). Lesions were present, usually at the leaf tips, and pycnidia were formed on the upper surface of the leaf on these lesions. Small reddish-brown necrotic areas were usually present further down the leaf blade. It is usually associated with wet conditions.

Fig. 3.—*Alternaria zinniae* on leaf of *Zinnia elegans*.
—*Photo: H. Powell.*

11. *Alternaria zinniae* Pape, Angew. Bot. *24:* 69; 1942.
On leaves and stems of *Zinnia elegans* Jacq., Mt. Albert, Auckland, J.M.D., Feb. 1956 (Fig. 3).

Lesions circular, 0.2-1 cm. diameter, becoming angular and fusing together; conidiophores formed on both surfaces of leaf,

singly or in clusters of 2-4, olive brown, septate, but unbranched up to 150μ long, 5-8μ wide. Conidia smooth, muriform, oval or cylindrical, terminating abruptly in a long beak, 3-8 transverse septa with 0-5 longitudinal septa, 20-64 $\times$ 6.5-12μ with beaks 10-15μ long. Reddish-brown necrotic areas were present both on leaves and stems; the dead tissue became very brittle, fragmented, and gave the plants a very ragged appearance. It has been recorded from Europe and North America; it is said to be seed carried and to be responsible for a seedling blight as well as a leaf spot.

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BLOOD CHEMISTRY AND HAEMATOLOGY IN SHEEP ON DECREASING LEVELS OF FEED INTAKE FOLLOWED BY STARVATION

By R. R. White, K. R. Christian, and V. J. Williams, Ruakura Animal Research Station, Department of Agriculture, Hamilton

(Received for publication, 16 August 1956)

Summary

Twelve wethers were individually penned for 128 days and fed dried spring grass of uniform composition. Daily feed intakes were decreased in steps from 1000g. to 300g. at varying intervals and to zero for the last 9 days. Live weight at the end of the 1000g. period averaged 107 lb. and at the end of the starvation period, 73 lb.

At intakes of 500g. or over, no marked changes were observed in blood samples examined at weekly intervals.

Successive lowering of intakes below 500g. resulted in no change in blood non-protein nitrogen and amino-acid nitrogen. Blood sugar, total keto-acids, and serum calcium showed successive slight falls. Blood total acetone bodies, inorganic phosphorus, and serum magnesium showed major changes on starvation.

The haematological picture showed no marked changes while intakes were greater than 400g. Further intake reductions produced considerable lowering of circulating eosinophils, and affected neutrophils, lymphocytes, and total leucocytes. Haemoglobin and haematocrit remained within normal limits.

The results are considered in relation to possible metabolic changes.

Introduction

Changes in blood constituents may reflect alterations in metabolism and therefore often yield valuable information on the underlying causes of pathological conditions. However, their interpretation may be difficult or misleading if the conditions are accompanied by loss of appetite which may itself bring about changes in blood constituents and it is important to know if and to what extent this occurs so that allowance may be made for it in pathological investigations. Although the effects of underfeeding and starvation on blood constituents of non-ruminants have been extensively studied, there have been few systematic experiments on underfed but otherwise normal ruminants.

The aetiology of the disease known in New Zealand as hogget ill-thrift is obscure (Clarke and Filmer[1]) and it was thought that an investigation of blood constituents might yield information by which it could be more closely characterized. The condition results in reduced growth rate during autumn and early winter, and as it may be accompanied by considerably reduced feed intake (Williams and Christian[2]) a preliminary trial has been carried out to follow blood changes produced by controlled underfeeding of healthy animals. Chemical blood constituents likely to detect alterations in carbohydrate, protein, fat, and mineral metabolism were selected for study. In addition, haemoglobin and haematocrit should indicate general nutritional status, and leucocyte responses reflect reaction to imposed stress.

EXPERIMENTAL

Twelve mature Romney wethers were individually penned for 127 days. Dried spring dairy pasture of uniform composition, consisting mainly of ryegrass, prairie grass, and white clover, was fed in two equal amounts each day for 76 days and thereafter in one feed each morning for 42 days. Partial chemical analysis of the feed is given in Table 1.

TABLE 1. PARTIAL ANALYSIS OF FEED

Dry matter = 85%	
Constituents as % of Dry Matter	
Crude protein	17.8
Crude fibre	21.5
Ether extract	9.6
Ash	10.6
Calcium	0.42
Magnesium	0.29
Phosphorus	0.19
Silica	2.4

An initial feed level of 1000g./day was chosen as a highly productive ration and was maintained for 32 days. Intake was then reduced to 800g. (5 days), 700g. (5 days), 600g. (12 days), and 500g. (8 days), to bring about gradual losses in live weight. Rations of 400g. and 300g. were maintained for 31 days and 24 days, respectively, to detect possible long-term effects of low feed intake. The animals were then starved for 9 days and finally returned to *ad lib.* feeding. Water was always available.

Each animal received 1 oz. phenothiazine at the start of the experiment and 250,000 I.U. of vitamin D as calciferol on the 46th day.

Live weights were measured at approximately five-day intervals.

Blood samples were collected from the jugular vein before the morning feed at weekly intervals and twice weekly toward the end of the trial. The blood was allowed to flow freely from a needle into each of four bottles:

1. Plain—serum calcium and magnesium.
2. Containing Long's preservative[3]—total keto-acids.
3. Containing oxalate and fluoride—all other biochemical determinations.
4. Containing balanced oxalate—haematological studies.

Analytical methods were as follows:

Blood Sugar. A modification of the method of Trevelyan and Harrison[4], using cooling and rapid mixing during addition of anthrone solution to filtrate obtained from barium hydroxide—zinc sulphate precipitation.

Blood Total Acetone Bodies. A method developed in this laboratory similar to that of Michaels *et al.*[5]. Colour production with salicylaldehyde.

Blood Total Keto-acids. Friedemann and Haugen[6].

Blood Inorganic Phosphorus. Briggs[7].

Blood Non-protein Nitrogen. White *et al.*[8]—Folin-Wu filtrate.

Blood Amino-acid Nitrogen. McDonald and White[9].

Serum Calcium. Clark and Collip[10].

Serum Magnesium. Denis[11]. Colour production as for phosphorus.

Haemoglobin. Thomas *et al.*[12]

Haematocrit. Wintrobe and Landsberger[13].

Total Leucocytes. Haemacytometer count on 1:20 dilution of blood.

Differential Leucocytes. Leishman's stain.

Eosinophils. Haemacytometer count using Manner's diluting fluid[14].

The animals were used coincidently for other investigations. Throughout the experiment they were bagged for the total collection of faeces, bags being changed twice daily. Each animal received a capsule of chromium sesquioxide night and morning for the first 75 days. Rumen samples were collected once a week for the first 11 weeks.

Results

Weight Changes and General Condition of Animals

Average live weights are shown in Fig. 1.

At the start of the experiment the animals were in forward store condition with an average live weight of 96 lb. On the 1000 g. level they rapidly fattened and reached a live weight of 107 lb. in 17 days. Reductions of intake to the end of the 500 g. level resulted in a loss of 11 lb. over 30 days. Further intake reduction to the end of the 300 g. level was accompanied by a loss of 14 lb. over 55 days. The animals were then thin, restless, and very hungry. Starvation resulted in a fall of 10 lb. live weight in 10 days. During this period the animals spent much time lying down but never failed to show interest when approached. Their wool became loose and tended to fall out, but otherwise they appeared healthy. On resumption of *ad lib.* feeding, appetites were low and erratic for two or three days and thereafter returned to normal with a marked response in live weight.

Average daily water consumption for the last eight days of the 300 g. period was 1160 g. and for the starvation period 460 g.

Blood Findings

Biochemical and haematological data are shown in Tables 2 and 3, respectively. For purposes of statistical analysis results obtained at the following feed levels were treated as separate groups; 1000 g.; 700-500 g.; 400 g.; 300 g.; zero. No samples were taken at the 800 g. level.

The following statistically significant changes in blood constituents were obtained:

TABLE 2. Blood Chemical Constituents (mg./100ml.)

Intake (g./day)	Sampling Day	Sugar		Ketone Bodies		Keto-acids		Amino-acid Nitrogen		N.P.N.		Inorganic Phosphorus		Serum Calcium		Serum Magnesium	
	—	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.
	15	44	8	1.3	0.2	—	—	9.4	0.6	39	3	5.1	0.6	10.7	0.5	2.5	0.4
	22	47	6	1.5	0.2	1.5	0.3	9.0	0.5	37	2	5.0	0.4	—	—	—	—
1000 (33 days)	26	47	5	1.5	0.2	1.6	0.2	9.1	0.4	—	—	5.1	0.7	10.7	0.4	2.5	0.1
	28	—	—	—	—	—	—	—	—	—	—	—	—	10.6	0.4	2.6	0.3
	33	43	6	1.3	0.2	1.8	0.2	9.2	0.4	36	2	5.2	0.5	10.7	0.4	2.5	0.2
	Av.	**45**	**6**	**1.4**	**0.3**	**1.6**	**0.3**	**9.2**	**0.5**	**37**	**2**	**5.1**	**0.5**	**10.7**	**0.4**	**2.5**	**0.3**
800 (5 days)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
700 (5 days)	40	43	4	1.5	0.3	1.6	0.1	9.8	0.6	36	2	4.8	0.5	11.3	0.3	2.8	0.2
	47	46	5	2.3	0.3	1.9	0.2	9.7	0.5	35	2	4.7	0.7	11.1	0.5	2.4	0.2
600 (12 days)	54	45	5	1.6	0.3	1.7	0.3	9.2	0.7	36	2	4.6	0.5	11.1	0.5	2.6	0.2
500 (8 days)	61	44	4	1.7	0.2	1.6	0.2	8.6	0.8	36	3	4.5	0.6	11.0	0.4	2.5	0.2
	Av.	**44**	**5**	**1.8**	**0.4**	**1.7**	**0.2**	**9.3**	**0.8**	**36**	**2**	**4.7**	**0.6**	**11.1**	**0.4**	**2.6**	**0.2**
400 (31 days)	68	41	3	1.8	0.3	1.9	0.3	8.6	0.8	34	3	4.7	0.7	10.9	0.6	2.8	0.2
	75	40	4	1.2	0.2	1.5	0.3	9.1	0.8	37	3	4.6	0.8	10.8	0.7	2.7	0.2
	91	40	4	1.2	0.2	1.3	0.5	9.9	0.8	38	3	4.9	0.6	10.7	0.6	2.5	0.2
	Av.	**40**	**4**	**1.4**	**0.4**	**1.6**	**0.1**	**9.2**	**0.9**	**36**	**3**	**4.7**	**0.7**	**10.8**	**0.6**	**2.7**	**0.2**
	96	40	4	1.6	0.4	1.4	0.5	9.4	0.7	39	3	5.0	0.7	10.7	0.6	2.6	0.1
	100	38	5	2.0	0.9	1.5	0.5	10.0	0.6	37	2	4.9	0.8	10.4	0.7	3.0	0.3
300 (24 days)	105	40	4	1.6	0.4	1.5	0.4	9.7	0.7	38	2	5.0	1.1	10.3	0.8	2.5	0.1
	111	36	5	1.3	0.4	1.3	0.4	—	—	37	3	4.5	0.7	10.6	0.7	2.4	0.2
	118	38	2	1.7	0.5	1.1	0.2	10.7	0.9	44	3	4.8	0.7	10.2	0.7	2.4	0.2
	Av.	**38**	**4**	**1.6**	**0.6**	**1.4**	**0.4**	**10.0**	**0.8**	**39**	**4**	**4.8**	**0.8**	**10.4**	**0.7**	**2.6**	**0.3**
	121	31	5	7.1	1.3	1.2	0.3	9.8	0.5	43	5	7.0	1.1	9.4	0.8	2.2	0.2
Nil (9 days)	124	35	5	8.1	1.3	1.2	0.4	11.5	1.3	37	4	7.6	1.2	8.9	0.9	1.7	0.2
	128	32	8	9.0	2.0	1.1	0.1	10.3	0.6	34	4	8.2	1.0	9.1	0.6	1.8	0.2
	Av.	**33**	**6**	**8.1**	**1.4**	**1.2**	**0.3**	**10.7**	**1.4**	**38**	**6**	**7.6**	**1.3**	**9.1**	**0.8**	**1.9**	**0.3**

TABLE 3. Haematological Findings

Intake (g./day)	Sampling Day	Leucos. × 10^3 per cu.mm.		Neutros. % of Leucos.		Lymphos. % of Leucos.		Eosinos per cu.mm.		Haemoglobin g./100 ml.		Haematocrit r.b.c. vol. %	
		mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.
1000 (33 days)	15	9.9	2.7	13	6	78	12	1040	1030	11.7	0.9	42	3
	22	10.7	3.0	13	9	80	12	780	760	12.2	0.9	40	3
	26	10.4	2.7	20	8	68	15	1190	680	12.1	0.5	36	3
	33	11.0	1.9	17	8	71	11	1180	610	12.3	0.7	37	3
	Av.	**10.5**	**3.1**	**16**	**8**	**74**	**13**	**1050**	**800**	**12.1**	**0.8**	**39**	**4**
800 (5 days)	—	—	—	—	—	—	—	—	—	—	—	—	—
700 (5 days)	40	10.6	3.0	17	6	71	8	1240	890	12.0	0.7	38	3
	47	9.5	2.2	13	6	73	12	1290	1000	11.2	0.5	37	3
600 (12 days)	54	9.6	2.9	13	4	76	7	990	620	11.0	0.4	36	2
500 (8 days)	61	10.0	2.2	15	8	72	10	1100	700	10.9	0.5	36	3
	Av.	**9.9**	**2.6**	**14**	**6**	**73**	**9**	**1160**	**810**	**11.3**	**0.7**	**37**	**3**
400 (31 days)	68	9.6	2.3	14	4	75	9	940	630	11.0	0.7	36	3
	75	8.6	2.6	16	5	71	7	780	310	10.8	0.6	35	3
	91	9.9	2.5	15	9	77	12	650	280	11.5	0.5	36	3
	Av.	**9.4**	**2.5**	**15**	**7**	**74**	**9**	**790**	**450**	**11.1**	**0.7**	**36**	**3**
300 (24 days)	96	8.8	2.4	14	7	80	8	580	360	11.5	0.7	37	3
	100	9.1	2.4	18	7	73	10	570	320	11.3	0.4	35	3
	105	9.0	2.0	17	7	76	10	550	360	10.2	0.5	33	3
	111	8.0	1.5	23	20	73	22	470	240	9.9	0.3	32	2
	118	8.2	2.0	20	15	76	14	400	220	9.8	0.2	32	2
	Av.	**8.6**	**1.6**	**18**	**12**	**76**	**14**	**520**	**310**	**10.5**	**0.8**	**34**	**3**
Nil (9 days)	121	6.3	2.1	24	9	70	10	250	180	9.8	0.2	34	3
	124	5.6	2.1	28	12	66	13	260	130	11.4	0.9	34	4
	128	5.8	1.4	31	8	65	9	170	120	11.4	0.7	37	3
	Av.	**5.9**	**1.9**	**28**	**11**	**67**	**11**	**230**	**150**	**10.6**	**0.9**	**35**	**4**

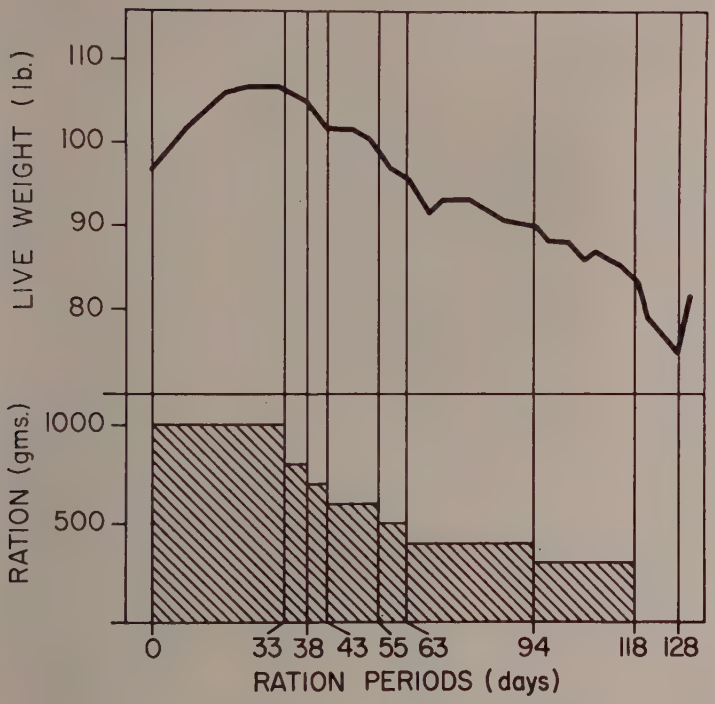

Fig. 1.—Changes in feed intake levels and liveweights.

Biochemical

Blood Sugar: successive decreases on lowering intake from 500 g. to 400 g., from 400 g. to 300 g. and from 300 g. to zero.

Total Acetone Bodies: a minor temporary rise following initial feed reductions and a major rise on starvation.

Total Keto-acids: a considerable fall between 500 g. and starvation.

Non-protein Nitrogen: none.

Amino-acid Nitrogen: none.

Inorganic Phosphorus: on first lowering of feed intake a slight fall to a level which was maintained until starvation when a major rise took place.

Serum Calcium: a rise following initial lowering of intake, a corresponding fall during the 400 g. period, a further slight fall at the 300 g. level and a greater fall on starvation.

Serum Magnesium: none while the animals were fed. A fall during starvation.

Haematological

Total Leucocytes: a slight fall between the 700-500 g. and the 300 g. levels, and a further substantial decrease during starvation.

Neutrophils, per cent of Leucocytes: none while the animals were fed. A rise during starvation.

Lymphocytes, per cent of Leucocytes: none while the animals were fed. A fall during starvation.

Eosinophils: successive large falls on lowering feed intakes from 500 to 400 g., from 400 to 300 g. and from 300 g. to zero.

Haemoglobin: a slight but steady fall during reduction of intake from 700 g. to 300 g. No change on starvation.

Haematocrit: a fall between the 700-500 g. level and the 300 g. level.

Discussion

Metabolic changes can not be described in detail from a consideration of blood constituents alone, but certain features of this study are sufficiently clear-cut to make interpretation possible. Although considerable alterations in energy metabolism must have resulted from reduction of feed intake from 1000 g. to 500 g./day there were no marked blood changes. The slight fall in phosphate and temporary rise in ketone bodies that occured in the early stages of the experiment may have been due to altered metabolism but neither constituent showed abnormal values. The fall in blood sugar produced by reduction of intakes to 400 g./day must have resulted from a deficiency of dietary energy. Allcroft and Strand[15], Groenewald *et al.*[17] and Reid[18] have reported only moderate hypoglycaemia in non-pregnant sheep fasted for various periods. The hypoglycaemia reported in this experiment was not severe at this stage but is considered to be of biological importance as it was accompanied by changes in leucocytes, particularly the fall in eosinophils, commonly used as indications of reaction to stress (Dalton and Selye[19]. This eosinophil change was greater than could be attributed to the normal, very large variability. Engel[20] suggested that carbohydrate and protein deficiency may constitute a form of stress, while Alexander and Ash[21] showed that hypoglycaemia in the horse is accompanied by eosinopenia. Wynne[22] found complete absence of circulating eosinophils and evidence of adrenal hyperactivity in a sheep dying of starvation.

If energy deficiency is indeed a form of stress, adaption to it should result in the utilization of tissue protein for energy production (Long *et al.*[23]). At the 400 g. and 300 g. levels, lack of any increase in ketone bodies showed that augmentation from this source was sufficient to ensure the efficient utilization of depot fat. Blood amino-acid and non-protein nitrogen gave no direct evidence of utilization of tissue protein.

During the starvation period, when all energy must have been obtained from body reserves, blood sugars fell to the lowest limit of reported normality and there was strong reaction to stress, as shown by very low eosinophil levels and a change in the lymphocyte: neutrophil ratio. The consequent utilization of tissue protein did not supply sufficient glycogenic fatty acid for the complete oxidation of depot fat, and a considerable rise in ketone bodies occured. The increased blood phosphate was probably a reflection of increased utilization of tissue protein. Dale *et al.*[24] ascribed to the same cause a marked rise in urinary phosphate in fasting cattle, although they found no increase in blood phosphate. Lack of any rise in amino-acid or non-protein nitrogen during this period in the present experiment can only be assumed to be due to efficient urinary excretion of nitrogenous end-products.

The fact that keto-acids did not rise during the experiment, but showed a regular fall, indicates that there was no deficiency of vitamin B. The change observed may have been a reflection of a progressive lowering of metabolic rates.

Lowering of the haemoglobin and haematocrit during the period of feed reduction could possibly be ascribed to either protein or iron deficiency, or both, but levels were at no time abnormally low. Lack of further depression on starvation could have been due to haemo-concentration brought about by the considerable fall in water consumption.

Serum calcium and magnesium were probably affected by alterations in general metabolism as well as by dietary deficiency and it is of interest that, although magnesium did not alter until the starvation period, calcium showed a temporary slight rise in the initial stages of lowering of intake. This could have been due to the coincident lowering of phosphate. Both calcium and magnesium were well within normal limits at the lowest intake level and fell only to the lower limits of normality during starvation. Probably values at this stage were affected by the marked rise in phosphate, although Dale *et al.* who also reported falls in serum calcium and magnesium in their fasting cattle, found no increase in blood phosphate. However, their animals had been suddenly deprived of feed, and, moreover, the pre-starvation calcium figures reported by them were very high by generally accepted standards and fell only to normal levels.

Our results show that large reductions in feed intake may occur before major changes appear in the blood constituents studied. In applying them to investigations on disorders accompanied by low feed intake, consideration must be given to the probability that, because of their greater energy requirements, grazing animals would show blood changes at higher levels of intake than penned animals. Furthermore, the degree and rate of fall of intake in certain pathological disorders would be considerably different from that imposed in the present experiment, in which, because of the long depletion period, body reserves were probably very low toward the end, although reasonable opportunity was given for animals to adapt themselves to each feed level. In disorders accompanied by very rapid fall in intake, blood changes might be quite different from those reported here, and further investigations, covering other blood constituents and other degrees and rates of intake reduction, would be of value.

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EXCESS SOLUBLE SALTS IN GLASSHOUSE TOMATO SOILS

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Summary

Osmotic pressures of saturation extracts of glasshouse tomato soils associated with healthy and affected plants have been correlated with (1) weight of salts from (a) 1 part soil to 5 parts water and (b) saturation extracts (2) conductivities of such extracts and (3) osmotic pressures calculated from the composition of these extracts. Effects of ratio of soil to water on the amounts of dissolved ions have been examined over a wide range.

Conclusions were as follows:

(1) Conductivity determinations provide a more reliable estimate of osmotic effects than weight of salts extracted.

(2) Conductivity of the saturation extract provides a convenient and reasonably accurate measure of osmotic effects. Because of the presence of calcium sulphate in these soils, saturation extracts are much more reliable than extracts with a wider ratio of soil to water.

(3) The critical conductivity of the saturation extract is 6 to 7 millimhos per cm.

(4) As the ratio of soil to water is decreased, calcium from calcium sulphate may exchange for potassium, magnesium, and sometimes sodium, and increase the amounts of these ions in solution.

(5) Analyses of the water extracts indicate that the fertilizers mainly responsible for reduced yields from excess soluble salts are potassic and nitrogenous compounds.

Introduction

Very heavy fertilizer applications are frequently used in growing glasshouse tomatoes. In outdoor soils exposed to rain, excess soluble ions or unwanted residues are washed out, but in glasshouse soils such leaching will occur only when surplus water is applied. Overfertilized, or insufficiently washed, glasshouse tomato soils are common throughout New Zealand.

The excess fertilizers or residues result in a disorder which is essentially that of physiological drought. Details of the nature of the problem, visual symptoms, and practical methods of control have been described in a general article[1]. Weight of crop harvested may be seriously reduced through reduction in number and size of fruit. Symptoms resemble those due to drought except that wilting is not always a characteristic symptom. Usually, affected plants adapt themselves to the water deficit by becoming very "hard" and stunted, with dark foliage, small leaves, and spindly stems.

Uptake of water by plants is affected by (a) soil moisture tension and (b) osmotic pressure of the soil solution. American work with culture solutions[2] indicates that the effects are additive, while recent work[3] suggests that the critical combined stress for tomatoes is 4 to 5 atmospheres. In commercial glasshouse culture in New Zealand, it is normal practice to keep the soil relatively dry for the first 6 to 8 weeks

after planting out. Even during this period the soil-moisture tension typically rises to only half to one atmosphere, hence it is evident that the dominant effect in this problem is osmotic pressure of the soil solution. This paper discusses particularly some problems in the analytical diagnosis of excess soluble salts.

When analyses for soluble salts in glasshouse soils were commenced in 1949 as part of the Department's advisory service to growers, a 1:5 ratio of soil to water was employed for the preparation of the extracts, and the conductivity of the resulting suspension was determined at 25°C. (For reporting to growers, values were converted to "per cent soluble salts" in the air-dry soil, by applying a factor of 300, the value found applicable for certain saline soils of marine origin). It was soon realized, however, that there are serious limitations to this technique as a measure of osmotic effects because of (a) variable and frequently very high amounts of calcium sulphate in glasshouse soils and (b) differences in moisture-holding properties of different soils. The presence of the slightly soluble calcium sulphate can result in a gross overestimate of the osmotic pressure of the soil solution when a wide ratio of soil to water is used. Furthermore, osmotic effects are a function of the concentration of salts in the soil solution rather than the concentration in a given weight of dry soil. Reliable analytical methods should therefore measure osmotic pressures of the soil solutions or closely related properties.

A detailed study was commenced in 1952 to assess the reliability of the values obtained by available rapid methods and to establish suitable diagnostic criteria. Ideally, analyses should be made on the solution actually in contact with the soil particles under normal field moisture conditions. The separation of the soil solution can be accomplished by displacement methods such as described by Bird and Martin[4] or by expressing the moisture by means of pressure membrane equipment[5]. Such methods, however, are inconvenient as they are time consuming and require large samples to provide sufficient solution for analysis by physical or chemical methods. Extracts at moisture saturation provide a practical compromise. In this study the osmotic pressures of saturation extracts, determined from freezing point depressions, have been correlated with (a) weight of salts from 1:5 and from saturation extracts (b) conductivity of such extracts and (c) osmotic pressures calculated from the composition of these extracts. Effects of different ratios of soil to water on the amounts of dissolved ions have also been examined.

Methods

Field sampling: Most samples were submitted by horticultural instructors and were taken within the plant row to 9 in. depth. Special samples at different depths were taken from local glasshouses on the Horotiu sandy loam soil type to follow seasonal trends in levels and effects of watering. Despite the presence of calcium carbonate in some of the soils, their pH values (1:2½ soil:water suspension) fell in the range 5.1 to 6.9, mostly between 5.7 and 6.5.

Moisture: Air-dried soils were used and moisture contents determined by oven drying at 100°C. for 24 h. Approximate moisture-equivalent (M.E.) values were determined by the Bouyoucos method[6], with suction adjusted to one-third atmosphere, where necessary by compacting the soil in the moist condition during suction.

The moisture condition is considered by growers to be correct for planting out when the soil just holds together when lightly squeezed in the hand. "Minimum" moisture levels were determined on soils brought just to this condition by adding water as a fine spray to the partly dried soil and thoroughly mixing. (A more precise measure of the minimum levels used may be possible when soil moisture tensiometers are more generally used). Soil-moisture tension at the minimum moisture level was determined for several soils by reference to soil-moisture retention curves determined with the pressure membrane equipment.

Extraction: For 1:5 extracts, 20 to 50 g. air-dry soil were shaken for 1 hour with 5 times the weight of distilled water, and the suspension used for conductivity measurements. For detailed analyses the supernatant liquid was filtered through a fine filter paper till clear and stored for the minimum time at about 5°C. For saturation extracts approximately 200 g. soil were taken. The criteria of saturation as set out in U.S.D.A. Handbook No. 60[2] were used, and, after the saturated soils were left to stand overnight, the extracts were filtered off under suction.

Conductivity: Specific conductances (κ) were determined at 25°C. by means of an A.C. bridge with cathode-ray visual null indicator (electric eye). (κ is conductivity of 1:5 extracts, κ_s of saturation extracts).

Analytical: Nitrate, phosphate, bicarbonate, and chloride were determined by standard methods, sulphate by Steinbergs' turbidity method[7], ammonium by semi-micro Kjeldahl distillation, potassium, sodium, and calcium by flame photometry (calcium after removal of aluminium and phosphate), and magnesium spectrographically. Nitrate was determined as soon as possible after extraction, as values were found to decrease with time. Results were calculated to oven-dry soil basis.

Osmotic pressures (*O.P.*): Freezing point depressions were converted to osmotic pressures using the formula $12.06\Delta T - 0.021\Delta T^2$. Osmotic pressures were also calculated from the composition of the extracts using graphs prepared from data in volume 4 of International Critical Tables[8]. For comparison of relative contributions by different salts, conductivity values were calculated at a given osmotic concentration, by reference to the above and to data in volume 6[9]. The values are summarized in Table 4 which is an extension of the table prepared by Magistad and Christiansen[10].

Results

In Table 1 a comparison is made of the composition of 1:5 and saturation extracts of several soil samples adjacent to affected and healthy plants, each pair of samples being taken from the same glasshouse. These examples show typical analytical figures for the different constituents, the sums of the ions measured, and the osmotic pressures of the dissolved salts calculated (a) from freezing-point depressions and (b) from the compositions. For the latter calculations a constant calcium-sulphate solubility of 0.208% $CaSO_4$ at 25°C. was assumed, and an osmotic pressure of 0.5 atmosphere for the saturated solution, the value found from freezing-point depression for a saturated solution of gypsum in distilled water. Bracketed figures are doubtful and there was insufficient sample available for check analysis. "Tr" means less than 0.001%. $\kappa \times 10^3$ values for 1:5 extracts have been corrected for moisture in

TABLE 1. COMPARISON OF SOILS (0-9 in.) ADJACENT TO AFFECTED AND HEALTHY PLANTS

Soil	% Moisture		Soil : Water	Grammes per 100 g. oven-dry soil												$CaSO_4$ Satn. %	Conductivity $\kappa \times 10^3$ at 25°c.	Osmotic Pressure (atmospheres)			
																		From F.P.		From Composition	
	M.E.	Min.	Extract	% H_2O	Ca	K	Na	Mg	NH_4	SO_4	NO_3	Cl	PO_4	HCO_3	Sum			At satn.	At min.	At satn.	At min.
A. Auckland	34	28	1:5	500	.390	.055	.028	.015	.001	.710	.460	.041	.005	.010	1.715	98	3.77	6.1	11.0	5.9	10.9
Affected			Satn.	53	.162	.032	.026	.011	Tr.	.079	.434	.041	Tr.	.005	0.790	100	16.3	6.07	11.2	6.0	10.8
Healthy			1:5	500	.265	.013	.015	.015	.001	.630	.125	.015	.002	.024	1.105	82	2.40	3.2	5.7	2.3	3.8
			Satn.	54	.073	.006	.010	.007	Tr.	.090	.142	.012	Tr.	.007	0.347	100	7.10	2.27	3.9	2.4	4.2
B. Ch'ch.	34	29	1:5	530	.238	.238	.028	.024	.005	.588	.231	.175	.021	.017	1.565	78	4.13	7.7	15.5	6.8	13.6
Affected			Satn.	60	.108	.150	.018	.014	.004	.127	.225	.169	.002	.008	0.825	100	16.6	5.91	11.7	6.0	12.0
Healthy			1:5	530	.117	.114	.014	.024	.002	.371	.112	.062	.016	.020	0.852	39	2.16	2.75	5.7	3.3	7.1
			Satn.	67	.070	.066	.010	.010	.001	.133	.103	.063	.001	.010	0.467	100	8.17	2.65	5.5	2.8	5.8
C. Ch'ch.	25	14	1:5	512	.115	.097	.012	.005	.002	.360	.034	.049		.025	0.699	38	1.70			3.8	10.1
Affected			Satn.	41	.034	.045	.009	.002	.001	.093	.037	.045		.010	0.276	100	7.91	2.53	6.4	2.4	6.0
Healthy			1:5	512	.088	.081	.009	.004	.004	.30	.004	.037		.016	0.550	29	1.36			3.2	8.5
			Satn.	41	.029	.033	.006	.001	Tr.	.095	.004	.032		.010	0.210	100	5.83	1.79	4.4	1.7	4.1
D. Greymouth	38	25	1:5	528	.114	.100	.031	.004	.022	.396	.081	.104	.008	.010	0.870	37	2.22	5.8	11.8	4.2	8.3
Affected			Satn.	53	.068	.066	(.037)	(.009)	.013	.131	(.098)	.112	Tr.	.004	0.538	100	12.6	4.37	8.7	4.6	9.2
Healthy			1:5	528	.057	.047	.014	.005	.004	.164	.071	.035	.009	.003	0.414	21	1.16	2.02	3.8	2.3	4.4
			Satn.	54	.047	.023	(.017)	(.007)	.002	.101	(.061)	.036	Tr.	.003	0.297	100	6.37	2.05	3.8	2.3	4.4
*E. Hamilton**	46	35	1:5	542	.289	.162	.030	.050	.004	.772	(.384)	.051	.003	.033	1.780	95	3.97			4.4	10.3
Affected			Satn.	88	.129	.107	.025	.042	.002	.429	.488	.043	.001	.005	1.271	100	11.7	4.77	11.2	4.4	10.4
Healthy			1:5	542	.230	.096	.023	.053	.006	.677	.269	.028	.004	.011	1.397	75	3.14			3.8	7.9
			Satn.	80	.098	.060	.015	.032	.003	.198	.260	.024	.001	.004	0.695	100	9.43	3.26	6.8	3.3	6.8
F. Wellington	34	18	1:5	500	.232	.083	.016	.022	.002	.445	.390	.043	.020	.011	1.264	61	2.76	7.5	22.0	5.4	15.5
Affected			Satn.	55	.132	.044	.014	.019	.001	.104	.386	.044	.002	.005	0.751	100	14.4	5.16	14.7	5.2	14.8
Healthy			1:5	500	.093	.043	.008	.010	.002	.235	.130	.011	.021	.007	0.560	30	1.40	2.65	7.4	2.4	6.7
			Satn.	58	.053	.016	.004	.007	.001	.105	(.115)	.008	.003	.002	0.314	100	5.27	1.69	4.3	1.7	4.2

* Results may be abnormally high as soil taken only to 6 in. depth and from ridged rows.

TABLE 2. Correlations of Results of Measurements on 32 Soils

Soil	1	2	3	4	5	6	7	8	9	10	11	
	% Moisture			Conductivity		Sum Ions (%)		Osmotic Pressures at Saturation				Ca SO_4% Satn. in 1:5 Ext.
										Calculated from composition		
	Satn.	M.E.	Min.	κ Satn. Ext.	κ 1:5 Ext.	Satn. Ext.	1:5 Ext.	From F.P.	From κ_s	Satn. Ext.	1:5 Ext.	
1	86	37	26	1.34	0.43	0.083	0.196	0.37	0.14	0.3	0.8	6
2	68	46	35	2.76	0.64	0.206	0.245	0.82	0.68	0.8	1.2	8
3	71	37	26	4.27	1.65	0.371	0.805	1.12	1.25	1.0	1.5	59
4	43	28	16	4.71	2.02	0.420	0.896	1.33	1.42	1.3	2.8	63
5	47	33	20	4.90	0.86	0.467	0.355	1.45	1.49	1.4	2.2	15
6	52	28	16	4.81	2.28	0.437	1.003	1.45	1.46	1.4	2.8	70
7	49	34	23	4.71	0.84	0.414	0.366	1.67	1.42	1.3	1.6	24
8	58	34	18	5.27	1.40	0.504	0.560	1.69	1.63	1.7	2.4	30
9	59	36	29	6.37	1.23	0.552	0.476	1.69	2.05	2.1	2.1	21
10	77	46	35	6.13	2.49	0.636	1.159	1.78	1.96	2.1	2.3	75
11	42	25	14	5.83	1.36	0.470	0.550	1.79	1.84	1.7	3.2	29
12	59	36	29	5.64	1.08	0.516	0.541	1.88	1.77	1.9	2.7	25
13	64	34	23	5.98	1.55	0.546	0.613	2.05	1.90	1.9	2.3	35
14	54	38	25	6.37	1.16	0.549	0.414	2.05	2.05	2.3	2.3	21
15	58	34	23	6.54	1.71	0.551	0.688	2.07	2.11	2.0	2.2	41
16	57	36	29	6.37	1.66	0.571	0.670	2.17	2.05	2.2	3.1	35
17	57	36	29	6.72	1.64	0.571	0.745	2.17	2.18	2.3	2.3	34
18	45	33	20	7.00	1.24	0.624	0.505	2.17	2.29	2.1	3.0	23
19	54	34	23	7.00	1.26	0.578	0.530	2.19	2.29	2.0	2.3	30
20	54	34	28	7.10	2.34	0.646	1.105	2.27	2.33	2.4	2.3	82
21	74	46	36	6.85	3.18	0.668	1.518	2.41	2.23	2.2	2.8	100
22	41	25	14	7.91	1.71	0.621	0.699	2.53	2.63	2.4	3.8	38
23	67	36	29	8.17	2.16	0.699	0.852	2.65	2.73	2.8	3.3	39
24	80	46	35	9.43	3.40	0.899	1.397	3.26	3.21	3.3	3.8	75
25	61	34	28	9.80	3.41	0.878	1.564	3.26	3.35	3.6	3.3	100
26	53	38	25	12.60	2.24	1.012	0.870	4.37	4.41	4.6	4.2	37
27	88	46	35	12.90	4.01	1.227	1.780	4.77	4.53	4.4	4.4	95
28	71	42	35	14.90	4.01	1.402	1.800	5.12	5.29	5.3	6.6	89
29	55	34	18	14.40	2.52	1.362	1.264	5.16	5.10	5.2	5.4	23
30	59	34	28	14.90	4.14	1.370	1.840	5.20	5.29	5.4	4.8	100
31	60	36	29	16.60	4.12	1.366	1.565	5.91	5.93	6.0	6.8	78
32	54	34	28	16.30	3.86	1.485	1.715	6.07	5.82	6.0	5.9	98
Mean	59.9	35.9	25.8	7.96	2.11	0.709	0.915	2.65	2.65	2.67	3.14	50
r	—	0.819	0.761	0.996	0.846	0.988	0.807	—	—	0.993	0.933	

air-dry soils. For soils A and F, the weight of soil taken for the 1:5 extracts was adjusted to allow for the moisture content of the air-dry soil.

Correlations of results of measurements on 32 soils are summarized in Table 2. The soils are arranged in order of increasing osmotic pressures (column 8). The sum of ions in the saturation extract is expressed as percentage in the solution; in the 1:5 extract as per cent of oven-dry soil. Values of the correlation coefficient r are for columns 2 and 3 compared with 1, and for columns 4 to 7, 10, and 11 compared with 8.

TABLE 3. Effect of Depth of Sampling and of Ridging and Watering on Levels in 1:5 Extracts

Date	Depth (inches)	Grammes per 100 g. air-dry Soil.										$CaSO_4$ Satn. %	$\kappa \times 10^3$ 25°c.
		Ca	K	Na	Mg	NH_4	SO_4	NO_3	Cl	PO_4	HCO_3	Sum		
1952 Season														
21 Oct.	0–1	.449	.237	.081	.100	.010	.540	1.640	.111	.002	002	3.172	68	8.83
	0–3	.174	.069	.009	.025	.003	.510	0.131	.010	004	.008	0.943	53	2.13
	3–6	.174	.084	.009	.016	.002	.525	0.133	.014	.001	.007	0.965	53	2.13
	6–9	.101	.109	.011	.009	.002	.349	0.115	.014	Tr.	.009	0.719	31	1.88
	9–12	.066	.117	.012	.004	.002	.266	0.099	.018	Tr.	.002	0.586	20	1.57
1953 Season														
11 Sept.	0–1	.284	.044	.019	.042	.002	.623	0.328	.039	.003	.019	1.403	79	3.37
	0–3	.139	.018	.013	.017	.002	.413	0.088	016	.004	.006	0.716	42	1.62
	3–6	.092	.020	.009	.009	.003	.266	0.049	.004	.003	.012	0.467	28	1.18
24 Sept.	0–1	.297	.040	.033	.035	.003	.671	0.342	.016	.003	.007	1.447	85	3.50
	0–3	.186	.023	.015	.030	.003	.548	0.122	.015	.004	.006	0.952	56	2.13
	3–6	.097	.021	.008	.015	.002	.300	0.054	.010	.004	.002	0.513	29	1.23
22 Oct.	0–1	.290	.044	.034	.053	.002	.821	0 374	.030	.003	.012	1.663	88	3.77
	0–3	.123	.018	.013	.018	.002	.401	0.072	.011	.005	.011	0.674	37	1.58
	3–6	.078	.018	.009	.010	.002	.241	0.035	.006	.004	.011	0.414	24	1.00
	6–9	.055	.019	.006	.005		.187	0.043	.004		.013	0.332	17	0.70
19 Nov.	0–3	.106	.013	.010	.014	.003	.307	0.058	.006		.022	0.539	32	1.22
	3–6	.110	.017	.009	.016	.002	.330	0.046	.006		.020	0.556	33	1.26
	6–9	.088	.022	.010	.014	.003	.265	0.040	.006		.017	0.465	27	1.04
20 Nov.	0–1	.376	.057	.058	.083	.007	.893	0.456	.043	.002	.022	1.997	113	4.46
	0–3	.055	.009	.007	.008	.003	.172	0.005	.001		.024	0.284	17	0.70
	3–6	.043	.011	.006	.005	.003	.142	0.010	.004		.024	0.248	13	0.58

Table 3 shows the effect of depth of sampling and of ridging and watering on levels in 1:5 extracts.

Fig. 1 to 4 show examples of the effect of different ratios of soil to water on the absolute amounts of ions dissolved in the solutions.

Discussion

Dominant Ions

General. Ca, K, SO_4, NO_3 are normally dominant (Table 1), but sometimes Mg, Na, and Cl concentrations are relatively high. Where appreciable amounts of sodium nitrate and potassium chloride fertilizers have been used, Na and Cl ions may predominate (not illustrated in the table). Usually NH_4, PO_4, HCO_3 ions are relatively unimportant.

Affected and Healthy Soils. Soils adjacent to affected plants show higher concentrations of total ions, and higher conductivities and osmotic pressures than comparable samples taken from alongside healthy plants (Table 1). The main difference in concentration of individual cations in saturation extracts in these pairs is in K (soils A, B, D, E, F) and in Ca (A, B, E. F), and among anions NO_3 (A, B, C, E, F), Cl (A, B, D, F), and sometimes SO_4 (E). Though SO_4 usually contributes a high proportion of the total osmotic pressure, in the examples quoted it is appreciably higher in the affected soils only in example E.

Comparison of 1:5 and Saturation Extracts. In all cases the ions in the 1:5 extracts total much more than in the saturation extracts. Ca and SO_4 are invariably much higher in the 1:5 extracts, while NO_3 and Cl are relatively constant, but the absolute amounts of K and sometimes Mg are also higher in the 1:5 extracts. In the examples cited in Table 1, these solutions show calcium-sulphate saturations (assuming 0.208% solubility) ranging from 21% to 98% and this means that the saturation extracts (requiring one sixth to one twelfth the quantity of water) are all saturated with calcium sulphate. The differences in total and individual ions are largely accounted for by increased solution of calcium sulphate in the 1:5 extracts compared with saturation extracts.

Reliability of Results from 1:5 and Saturation Extracts

Saturation extracts require about double the amount of water present in soils at minimum field-moisture levels found in practice, but, among all the tomato soils examined, 1:5 extracts use anything from 12 to 40 times as much water, depending on the moisture-holding properties of the soil. The average calcium-sulphate saturation of 1:5 extracts of typically fertilized affected soils has been found to be nearly 50% in the soils examined, with a range of 15% to 100%. If there is sufficient calcium sulphate present to saturate the 1:5 extract and no allowance is made for this, the calculated osmotic pressure at moisture saturation, for example 50% moisture, will theoretically be 4.5 atmospheres too high. If an average calcium-sulphate saturation of 50% is assumed, the maximum error will be approximately plus 2 atmospheres. If saturation extracts are used, however, the maximum error possible through ignoring dilution effects on calcium-sulphate solubility is only about 0.5 atmosphere. In practice this error can usually be eliminated, as all high-conductivity soils so far examined at moisture saturation (except two unusual examples where high sodium and chloride fertilizers were used) were saturated with calcium sulphate, thus allowing the application of a constant correction factor.

TABLE 4. Relation Between Osmotic Pressure, Conductivity and Salt Concentration for Different Salts

	Parts per million						Milliequivalents per litre						$\kappa \times 10^3$ at 25°c. (mhos per cm.)					
Atmospheres	0.1	0.2	0.5	1	2	4	0.1	0.2	0.5	1	2	4	0.1	0.2	0.5	1	2	4
$NaHCO_3$	175	350	880	1800	3750	7650	2.1	4.2	10.5	22	45	91	0.19	0.38	0.92	1.84	3.66	7.0
NH_4Cl	125	250	625	1260	2550	5200	2.3	4.7	11.7	24	48	97	0.34	0.67	1.65	3.23	6.22	12.4
NaCl	135	270	675	1360	2760	5620	2.3	4.6	11.5	23	47	96	0.29	0.56	1.37	2.69	5.25	10.3
KCl	170	340	860	1720	3530	7200	2.3	4.6	11.5	23	47	97	0.33	0.66	1.62	3.19	6.30	12.5
$CaCl_2$	175	350	880	1840	3720	7600	3.1	6.2	15.9	33	67	137	0.39	0.76	1.86	3.70	7.03	13.7
$MgCl_2$	150	305	765	1560	3150	6390	3.2	6.4	16.1	33	66	134	0.39	0.77	1.84	3.51	6.90	13.1
NH_4NO_3	180	360	920	1900	3770	7800	2.2	4.5	11.5	24	47	97	0.31	0.63	1.56	3.15	6 03	12.0
$NaNO_3$	200	400	1000	2000	4070	8250	2.4	4.7	11.8	24	48	97	0.29	0.55	1.35	2.61	5.00	9.6
KNO_3	235	470	1170	2400	4880	10100	2.3	4.6	11.6	24	48	100	0.32	0.63	1.57	3.14	6.10	12.0
$Ca(NO_3)_2$	270	545	1370	2780	5850	11950	3.3	6.6	16.7	34	71	146	0.41	0.79	1.89	3.83	7.60	14.2
$Mg(NO_3)_2$	250	505	1270	2550	5100	10350	3.4	6.8	17.1	34	69	140	0.39	0.77	1.85	3.83	7.20	13.7
Na_2SO_4	220	450	1170	2400	4970	10580	3.1	6.3	16.5	34	70	149	0.38	0.73	1.80	3.45	6.60	12.7
K_2SO_4	275	565	1450	3000	6150	13100	3.2	6.5	16.6	34	71	150	0.46	0.90	2.16	4.24	8.10	15.8
$CaSO_4$*	350	740	2080				5.1	10.9	30.5				0.51	0.98	2.21			
$MgSO_4$	315	665	1830	3970	8610	18700	5.2	11.0	30.4	66	143	310	0.51	0.97	2.28	4.15	7.60	14.0

* From measurements of F.P. depression and cross-reference to data in U.S.D.A. Handbook No. 60; pp. 104 and 157.

Results in Table 2 are consistent with the above evidence. Conductivity of the saturation extract (column 4) is highly significantly correlated with osmotic pressure of the same solution determined from freezing-point depressions (column 8) ($r = 0.996$). The relationship is O.P. $= 0.3787$ ($\kappa_s \times 10^3$) $- 0.360$ and the standard error of a computed value near the central point is 0.026 atmosphere. Values in column 9 (O.P. from κ_s) were obtained using this formula. Agreement between the pairs of figures is good, with no serious anomalies.

The concentration of ions in the saturation extract (column 6) also shows a good correlation ($r = 0.988$), and values are related to osmotic pressures (column 8) by the formula O.P. $= 4.1000$ (sum ions) $- 0.256$ with a standard error of the computed value of 0.044 atmosphere.

Osmotic pressures calculated from the analytical results for saturation extracts by reference to Table 4 show good agreement with measured osmotic pressures ($r = 0.993$) and the standard error of a value in column 10 as an estimate of column 8 is 0.035 atmosphere.

The relationship between measured osmotic pressures and the 1:5 values, though giving significant values of r (0.846 and 0.807 for conductivity and sum of ions respectively) are much poorer than for the saturation extracts, and the standard error of a computed value based on 1:5 extracts will be high. For example soils 5 and 6 in Table 2, with the same osmotic pressure of the saturation extract, show very different conductivities of the 1:5 extracts, and sums of dissolved ions. The calcium-sulphate saturation of the 1:5 extract of soil 5 was 15%, whereas for soil 6 it was 70%.

Another factor partly responsible for the poorer correlations with 1:5 extracts is the difference in moisture contents at saturation. In the examples in Table 2 the range was 41% to 88% (column 1). If allowance is made for these different moisture contents at saturation, by converting the measured osmotic pressures of the saturation extracts in column 8 to values at 50% moisture, a slightly better correlation is obtained in comparison of conductivities of the 1:5 extracts with these adjusted osmotic pressures ($r = 0.888$ instead of 0.846). A greater improvement is found when allowance is made for variable calcium-sulphate contents, by comparing osmotic pressures at saturation calculated from the composition of the 1:5 extracts (column 11) with observed osmotic pressures of the saturation extracts ($r = 0.933$ compared with 0.807 for the correlation using sum of ions in the 1:5 extract). The error of a determination, however, is still rather high and it is concluded that accurate results from 1:5 extracts cannot be expected, even if allowance is made for moisture-holding properties of the soils and calcium-sulphate content of the extracts. However, as the error is a positive one, low values for 1:5 extracts can confidently be interpreted as indicating low osmotic pressures of the soil solution.

Comparison of Conductivity and Concentration of Ions as a Measure of Osmotic Pressures

Figures in Table 4 show that in a variable mixture of salts there can be no exact simple relationship between weight of salts and osmotic pressure. For example a 0.14% solution of NaCl has the same osmotic pressure (1 atmosphere) as 0.17% KCl or 0.30% K_2SO_4 or 0.40% $MgSO_4$. On an equivalence basis it is seen that salts of which both

ions are monovalent have approximately the same osmotic effect. Salts with one divalent and two monovalent ions require about 50% higher equivalent concentration to give the same osmotic pressure, while the requirement for salts with both cation and anion divalent is nearly three times as high. On the other hand, the spread of conductivity values for a given osmotic pressure is less than for either weight or equivalence values. Conductivity should therefore be a more reliable criterion than either weight or equivalence of salts. Evidence from Table 2 is in accord with this. Correlation coefficients show that the relationship between conductivity and measured osmotic pressures is closer ($r = 0.996$) than between concentration of ions and osmotic pressures ($r = 0.988$). Determination of weight of extracted salts was discontinued because of the above considerations.

Factors Influencing Concentration of Soluble Salts

Effect of Fertilizer Rates. Probably the most important factor in determining build-up of soluble salts is rate of fertilizer applications, including side dressings. Troubles from excess salts have been associated especially with high rates of potassic and nitrogenous fertilizers. This has been discussed elsewhere[1].

Depth of Sampling. Table 3 shows results of seasonal tests on 1:5 extracts of samples taken from the relatively dry top inch of soil from rows ridged to approximately 3 in. depth, and 0.3 in., 3-6 in. and sometimes 6-9 in. and 9-12 in. samples taken from moist level soil adjacent to the plant ridges in a glasshouse in Hamilton where excess fertilizers had resulted in severe losses from soluble salts in 1952.

During the drying out of the soil, salts tend to move towards the drier zones and hence accumulate in the top layers of the soil. The gradients shown in Table 3 are a typical selection from a large number of analyses carried out on seasonal samples from this glasshouse over a number of years. Extracts were analysed in detail from this group only. Depth of sampling clearly influences the analytical results.

Effect of Watering on Conductivity. Flooding between the 1952 and 1953 crops reduced the conductivity. Before heavy watering was commenced in the 1953 season, the conductivity of 1:5 extracts of the 0-3in. layer on 22 October was 1.58 millimhos per cm., 1.00 at 3-6 in. and 0.70 at 6-9 in. After two moderate waterings the value fell to 1.23 in the top 3 in., but increased in the 3-6 in. zone to 1.26 and at 6-9 in. to 1.04. After very heavy watering, or partial flooding, on 19 November 1953 the values in the 0-3 in. and 3-6 in. zones fell to 0.70 and 0.58 millimhos per cm. respectively. Further results are not shown in the table, but continued heavy waterings up till the middle of December further reduced the value in the top 3 in. to 0.37, in the 3-6 in. and 6-9 in. zones to 0.27, and similar figures were found at 3 in. intervals down to 21 in., the lowest depth tested. A further set of samples taken in February 1954 after a long period of drying out, showed a rise in the value for the top 3 in. to 2.73, in the 3-6 in. zone to 0.67, in the 6-9 in. to 0.43, 9-12 to 0.40, while from 12 to 21 in. the value was 0.37 millimhos per cm. Rurther heavy watering before planting out in June 1954 lowered the value for the top 3 in. to 0.87, 3-6 in. to 0.50, and 6-9 in. to 0.23 millimhos per cm., and the figures have subsequently been maintained at similar levels up to the present day, with a normal

watering programme and using recommended moderate rates of fertilizers. Many other examples of the successful use of flooding to overcome trouble with excess soluble salts have been noted. In 24 such cases the average conductivity of 1:5 extracts of soil associated with poor crops was 2.35 millimhos per cm. before flooding, 0.68 afterwards, and subsequent crops were healthy. In normally watered houses, some seasonal fluctuations in conductivity values will automatically result from differential watering, the peak levels being found before heavy watering is commenced (usually about the fifth-truss stage) and at the end of the season before autumn flooding, especially where generous sidedressings have been used. Where the soil is free draining, the grower experiencing adverse effects of high soluble-salt concentrations can apply partial flooding and sucessfully reduce the osmotic concentration to safer levels while the plants are still growing. Soil samples taken *after* such thorough flooding will seldom correlate with incidence of stunting of growth or salt-induced blossom-drop. Frequently the results show the reverse of the expected trends as the grower, when advised of the remedial treatment, tends to apply much more water to the affected plants than to the healthy plants in the same house. Analyses are therefore best carried out during the periods of peak tests, that is before heavy watering is commenced, and the top 9 in. depth is suggested for level or trenched beds. Sidedressings may complicate the interpretation of results.

Effect of Watering on Concentration of Individual Ions. A comparison of concentrations of ions in the 0-3 in. zone on 19 November 1953 before flooding and 20 November after flooding indicates that the NO_3 ion is the most readily washed out, then Cl, while Ca, K, Na, Mg, and SO_4 are reduced to a less extent. Flooding is liable to reduce nitrate nitrogen below optimum concentrations, while reserves of potassium, phosphate, and other nutrients remain high. In the example quoted in Table 3, nitrate-nitrogen levels in the petioles of the leaves, measured in 2% acetic acid extracts, were 2,000 to 3,000 p.p.m. in the 1952 season and only one tenth these values a year later after flooding treatments and in the absence of further fertilizer applications. In some soils intensive watering may reduce potassium supplies to relatively low levels. In some glasshouses flooding to reduce salts has corrected magnesium deficiency induced by excessively high concentrations of ions, of which potassium appears to be the most important (see also Hunter[11]).

Effect of Ridging. In Table 3, values for the top 1 in. samples from relatively dry ridges are invariably much higher than for the moister or deeper zones. A type of "collar-rot" noted in some plants may be the result of plasmolysis by very high concentrations of salts in this zone. Secondary infection by *Rhizoctonia solani* was noted in one case. Raised pathways, especially when relatively dry, also accumulate salts. Both in ridges and pathways soluble salts are commonly present in the top 6 to 9 in. in amounts 2 to 6 times as high as in the moister plant beds. In sampling for analysis, such zones should be avoided or separately sampled. A similar effect of furrow irrigation of row crops in the field in saline soils has been discussed by Wadleigh *et al.*[12]

Of the individual ions, nitrate tends to be particularly concentrated in the top 1 in., while sulphate, relative to other ions, is lower in this zone. Nitrate appears to be the most mobile of the main ions, sulphate the least.

Critical Levels

It is generally accepted that growth is a simple function of the sum of soil moisture tension and osmotic pressure of the soil solution[2, 3, 13]. There is no threshold where a concentration of salts suddenly becomes toxic and precludes growth. Instead, the extent of growth reduction is roughly proportional to the osmotic pressure of the soil solution. Nevertheless, with increasing osmotic pressures, a stage must be reached when growth reduction becomes economically serious. In glasshouse tomato culture, the values associated with a reduction of about 10% in fruit yield can be considered as "critical levels". Bernstein and Pearson[3] concluded from their experiments with tomatoes in solution culture with fluctuating osmotic stresses from sodium chloride that the critical-stress value is approximately 5 atmospheres (4.6 to 5). Their term "critical-stress value" was not defined, but a yield reduction of approximately 20% is indicated from their data. With constant-stress treatments, growth was depressed 34.6% between 2 and 4 atmospheres, but fruit reduction would be much less. As there was no evidence of specific toxicities from the sodium and chloride ions, Bernstein and Pearson concluded that the effects on growth and fruiting were due solely to soil-moisture stresses. In glasshouse tomato culture in New Zealand, soil-moisture tension typically rises to $\frac{1}{2}$ to 1 atmosphere during the initial 6 to 8 weeks growth period when salt effects usually develop. Four atmospheres has therefore been taken as the critical value for the osmotic pressure of the soil solution.

1:5 Extracts. Some 22 pairs of samples associated with poor and healthy plants from the same or comparable houses showed average conductivities of 2.06 and 1.09 millimhos per cm. respectively on 1:5 extracts (0.62 and 0.33 "per cent salts" respectively). Six examples are shown in Table 1. In earlier work[1], approximate critical levels were worked out on the basis of the 4 atmospheres osmotic pressure value. Results were reported as "per cent salts" obtained by multiplying the specific conductance by 300. In Table 2 in the present work, the formula connecting conductivity of the 1:5 extract (column 5) with sum of ions (column 7) is $\kappa_{1:5} \times 10^3 = 2.1920$ (sum ions) + 0.1064. On a true weight basis the average conversion factor, using this formula, works out at 433. As already pointed out, however, these values can only be approximate averages, owing to the great variability in calcium-sulphate contents of tomato soils. In the interpretation of such values, moisture-holding properties of the soils must also be considered.

Saturation Extracts. Fifteen pairs of samples associated with poor and healthy plants showed average conductivities of the saturation extracts of 10.4 (range 6.9 to 16.6) and 5.1 (range 1.34 to 9.8) millimhos per cm. respectively. The osmotic pressures of 5 of these extracts quoted in Table 1 averaged 4.81 and 2.09 atmospheres respectively. Figures for soil E are omitted as abnormally high because the rows were ridged and sampling depth was 6 in. instead of the normal 9 in). At the "minimum" moisture levels—representing the true soil solutions at the important early-growth stage—the calculated values ranged from 6.4 to 14.7 (averages 10.5) atmospheres in the soil samples associated with affected plants, compared with 3.8 to 5.5 (average 4.4) atmospheres in relatively healthy samples from the same glasshouses. These results are consistent with the critical value of 4 atmospheres indicated by Bernstein and Pearson's work[3]. Some of the values for "healthy" samples may be too

high as the term "healthy" is relative and in badly affected houses such plants may still be stunted by high salts, though less seriously affected than the "unhealthy" plants. A further complicating factor is the fluctuating concentration and composition of the salts resulting from downward washing with heavy watering and sometimes upward movement between waterings. Where watering is much beyond field capacity, some of the salt components such as nitrates may be permanently washed out of the effective rooting zone, and if the soil sample has been taken after such intensive watering, the analytical results will be misleadingly low and unrelated to the incidence of symptoms of high salts, especially as some symptoms, such as blossom drop, will persist even though later plant growth may have returned to normal. There is local evidence, confirming earlier work of Walsh and Clarke[14], that high salts may cause permanent root injury and facilitate attack by root-rot organisms so that plants may remain unthrifty though salt concentrations have been reduced to safe levels.

The critical conductivity value for the saturation extract has been calculated as follows: As Wadleigh[15] has pointed out that peak tensions are likely to have a more marked effect on plant growth than mean tensions, "minimum" moisture levels have been used. Such moisture values obtained by the empirical technique were found to be roughly proportional to the saturation percentages ($r = 0.761$) (Table 2), and the validity of such a relationship is supported by Campbell and Richards' statement[16] that the saturation percentage is directly related to the field-moisture range. The average percentage moisture at saturation for samples in Table 2 was 59.9 and the average minimum 25.8. An osmotic pressure of 4 atmospheres at the minimum then corresponds to 2.075 atmospheres at saturation. In this calculation a simple proportionality has been assumed for the completely soluble components, after deducting 0.5 atmosphere for the saturated calcium-sulphate solution (the soil solutions both at the minimum and at moisture saturation were saturated with calcium sulphate). By substituting the value 2.075 atmospheres in the formula $O.P. = 0.3787\ (\kappa_s \times 10^3) - 0.360$, relating conductivity of the saturation extract to osmotic pressure, a critical conductivity value of 6.4 millimhos per cm. is obtained for the saturation extract (or $\kappa_s \times 10^3 = 6.4$ mhos per cm.).

U.S.D.A. Handbook No. 60[2] sets the figure of 10 millimhos per cm. as the conductivity value associated with a 50% decrease in yield of field tomato crops in saline soils. This figure is consistent with the above value of 6.4 for about 10% decrease in yield in glasshouse tomatoes.

Dunkle and Merkle[17] in experiments with tomatoes using high rates of mixed fertilizer (sodium nitrate, superphosphate, and potassium chloride) concluded that the "ceiling value" for 1:2 extracts (1 soil: 2 water) is approximately 2.27×10^{-3} mhos per cm. Serious reduction in growth was found above 3.00×10^{-3} mhos per cm. Grainger[18], from a glasshouse experiment with tomatoes, set a demarcation value of approximately 2.2×10^{-3} mhos per cm. (pC 2.65) for 1 soil : $2\frac{1}{2}$ water extracts.

The calculated critical conductivity value of 6.4 millimhos per cm. for the saturation extract is based on the assumption of $\frac{1}{2}$ to 1 atmosphere soil moisture tension over the early growth period. With the installation of soil moisture tensiometers, the tendency, if the makers' instructions

are followed, may be to work at lower initial tensions, up to a maximum of perhaps one-third atmosphere. If the tensiometer reading is not allowed to exceed 25 cm. mercury (⅓ atmosphere) the critical value will be higher, of the order of 7.5 millimhos per cm.

Data in Table 4 indicate that chloride solutions of a given conductivity value have higher osmotic pressures than sulphates, and sodium higher than potassium and especially calcium and magnesium. When sodium and chloride are relatively high, therefore, the critical conductivity level will be lower.

Further factors complicating the determination of critical levels or the interpretation of results include temperature effects on tolerance of high salts[19], and possible build-up of salts at the root surface. Richards

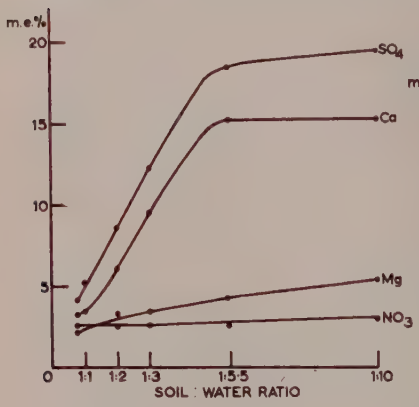

Fig. 1

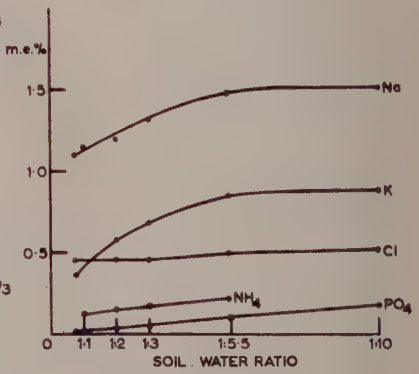

Fig. 2

Figs. 1 and 2.—Effect of ratio of soil to water on contents of ions in solution (soil F(a), Table 1).

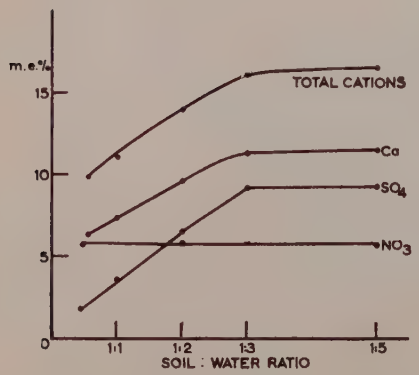

Fig. 3

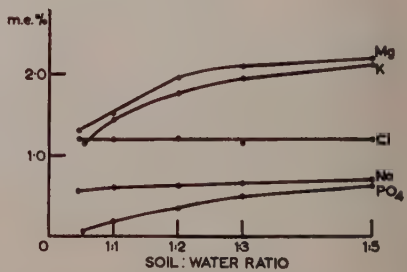

Fig. 4

Figs. 3 and 4.—Effect of ratio of soil to water on contents of ions in solution (soil 21, Table 2).

and Weaver[20] state that "the semipermeable characteristic of the plant root with its discriminating action against the uptake of most of the common salts must cause a build-up of the salt concentration at the root surface during moisture absorption". The lower tolerance at high temperatures may be due to a greater build-up of salts at the root surface with increased transpiration.

Effect of Ratio of Soil to Water on Absolute Levels of Ions Extracted

Fig. 1 and 2 (for F(a) in Table 1) show examples of changes in the amounts of ions in solution with different ratios of soil to water. Results are expressed as milliequivalents of the ions per 100 g. oven-dry soil.

The increase in total equivalents of cations paralleled the increase in SO_4 ions, from saturation through to 300% moisture content. Thereafter there was no further increase in Ca and SO_4 indicating that all the calcium sulphate was now in solution. Cl and NO_3 remained constant, Na increased slightly. The increase in the PO_4 concentration (quantitatively small) could have been due to increased solution of a slightly soluble phosphate. The increase in total cations and in sulphate ions from saturation to 300% moisture was approximately 7 milliequivalents per 100 g. oven dry soil, but the increase in calcium ions was only 5 milliequivalents. K and Mg, however, each increased by approximately 1 milliequivalent, indicating that at higher dilutions calcium has replaced potassium and magnesium, presumably from the exchange complex. The cation-exchange capacity of this soil (Waiwhetu sandy loam) was 19.8 m.e.%. Cations removed by leaching with neutral ammonium acetate were K 2.92, Na 0.46, Mg 1.46, and Ca 13.8 m.e.% respectively, while the corresponding cations in the 1 soil : 5 water extract were 1.06, 0.36, 0.77, and 4.5 m.e.% respectively. Soil 21 in Table 3 showed similar effects (Figs. 3 and 4). SO_4 increased by nearly 16 m.e.% but Ca by only 12. Here Mg (from heavy dressings of serpentine superphosphate and of ground serpentine) increased by 3 m.e.%, and K and Na each by approximately one half unit, giving a total of 4 m.e.%, the difference between the Ca and SO_4 equivalent increases. This soil was Horotiu sandy loam of cation-exchange capacity 32.5 m.e.%. Exchangeable bases in a recent sample from the same glasshouse were K 1.59, Na 1.45, Mg 3.35, and Ca 22.1 m.e.%.

Extracts of several samples at moisture equivalent, obtained by means of the pressure-membrane equipment, gave results which indicate that the trend of effects found at higher moisture contents extends down at least to moisture equivalent (approx. $\frac{1}{3}$ atmosphere tension).

Successively expressed fractions of the soil solution for soil 21 at saturation, obtained by the pressure-membrane method with increasing pressures rising in steps of 5, 15, 30, 45, and 60 lb. per sq. inch, and collecting at 0°c. to minimize evaporation losses, showed a progressive reduction in concentrations of salt components, including Cl and NO_3. Bower and Goertzen[23] have pointed out that this "negative absorption" effect is characteristic of inorganic salts in soil solutions. A similar effect on salt concentration, as indicated by conductivity measurements on the extracts, was noted by Reitemeier[21] at higher pressures, working with a field soil. It is therefore possible in the present work (and perhaps Reitemeier's) that the values at saturation may be fictitiously high because the actual solution analysed was obtained by means of a suction pump

and this solution is only the least strongly held fraction, which from the evidence of successively expressed fractions, may contain the highest concentrations of ions. Because of such effects, Reitemeier considered that soluble components such as chloride and nitrate should *decrease* on dilution, and in his work a reduction was noted in several soils examined, even over the range from saturation to 500% moisture. Changes of this kind were not detected in the present work.

CONCLUSIONS

(1) Because of the very variable composition of the extracts, conductivity values are more closely correlated with osmotic pressures than weight of salts.

(2) Results obtained with wide ratios of soil to water may be misleading because of different amounts of the slightly soluble calcium sulphate in the soil. The error diminishes as the ratio of soil to water approaches the actual conditions found in the field, consequently saturation extracts are much more reliable than 1:5 extracts.

(3) When a fixed ratio of soil to water is used, allowance must be made for the relative moisture-holding abilities of different soils. The use of saturation extracts automatically compensates for such differences.

(4) The conductivity of the saturation extract provides a convenient and reasonably accurate measure of osmotic effects.

(5) Factors influencing concentration of soluble salts include (a) rates of fertilizers (b) sidedressings (c) watering (d) depth of sampling (e) ridging or trenching of beds.

(6) For glasshouse tomato crops the critical conductivity of the saturation extract is about 6.4 millimhos per cm. when soil moisture tension is permitted to rise to nearly 1 atmosphere. The critical value will be higher, of the order of 7.5 millimhos per cm. at one-third atmosphere tension. Critical levels will be lower where sodium or chloride fertilizers have been used.

(7) As the ratio of soil to water decreases, calcium from dissolving calcium sulphate may partly exchange for potassium, magnesium, and sometimes sodium in some soils, resulting in an increase in the amounts of these ions in solution.

(8) In the soil moisture the cations that are appreciably higher in affected soils than in healthy are mainly potassium and calcium, sometimes sodium and magnesium, and the anions nitrate, chloride, and sometimes sulphate. It is concluded that the fertilizers mainly responsible for trouble from excess soluble salts are potassic and nitrogenous compounds, *not* superphosphate, as normally assumed[22].

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THE VITAMIN CONTENT OF NEW ZEALAND BUTTERFAT

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Summary

Vitamin A, carotene, xanthophylls, tocopherol, and iodine value have been determined at weekly intervals in factory butters from October to April.

The seasonal trends previously reported for vitamin A, carotene, and iodine value have been confirmed, and similar and closely correlated changes have been shown to occur in the levels of tocopherol and xanthophylls.

When total daily secretion of these substances, rather than concentration in the fat, is considered, seasonal trends are still apparent.

The concentration of carotene in the pasture remains high throughout the year, and the lower vitamin A potency of the milk fat over the summer months must result from a decreased availability rather than a lowered intake of carotene.

INTRODUCTION

The vitamin A potency of New Zealand butterfat was first investigated by Barnicoat[1] during the 1935–6 season. Under the conditions of all-the-year-round pasture feeding obtaining in New Zealand, the complicating effects due to stall-feeding on relatively low-carotene rations are eliminated and a uniformly high vitamin A potency might be expected for our butterfat. Barnicoat, however, found marked seasonal fluctuations in both carotene and vitamin A, the variations being in the opposite direction to those reported in the literature for European and American butters[2, 3]. A more extensive investigation covering the main dairying districts and extending over several seasons by McDowell and McDowall[4] confirmed Barnicoat's findings, and it is apparent that the potency of our butterfat decreases from a maximum of about 50 I.U./g during the late winter and early spring (July–October) to a minimum of about 30–35 I.U./g in late summer (February), with a subsequent increase through the autumn. Since the milk fat of cows calving in autumn follows the same trend as those calving in the spring, it is concluded that the changes in potency are seasonal rather than lactational[1, 5]. Similar fluctuations have been reported for a number of districts in Australia where dairying conditions are comparable to those existing in this country[6].

Since the seasonal fluctuations apparent in European and American butterfat depend largely on variations in the carotene intake of the animals[7, 8], Barnicoat suggested that the relatively low New Zealand summer potency might be due to a decreased carotene content of the pastures following the spring flush. However, subsequent work[9] has shown that the carotene content of our pastures is normally high throughout the year, giving grazing cows a daily carotene intake of 4–8 g and it has been pointed out that the minimum daily intake of 1.5 g, experienced only under abnormally dry conditions, is still considerably in excess of the level considered by Wilbur, Hilton, and Hauge[10] to be adequate for the production of milk fat of maximum vitamin A potency.

Further, a supplement of carotene in oil during mid-summer failed to raise the potency of the fat produced by pasture-fed cows[9], and it would seem therefore that the relatively low vitamin A potencies must result from a decreased utilization of carotene from summer pastures rather than from a deficiency of carotene in the diet.

McDowell and McDowall[4] were able to establish a close correlation between vitamin A potency and iodine number of the butterfat, and it seemed possible that the problem over the summer months was one of poor utilization from the pasture not only of carotene but of fat and fat-soluble substances. So far, observations had been limited to vitamin A and carotene, and the present investigation was therefore undertaken to determine whether the other related milk-fat components—tocopherol and xanthophylls—varied in a similar way. Further, weather records were followed in an attempt to determine any effect of climate on the vitamin A potency of milk fat either through a direct effect on the animal or indirectly through the pasture. Yield of milk fat was also considered, since this might be expected to influence the potency of the milk fat.

Experimental

Butter samples were obtained at weekly intervals throughout the 1952–53 season from a local butter-factory immediately after churning. The samples were taken from the same churning each week and were therefore as far as possible representative of the same group of cows. As has already been pointed out[4], repeated sampling of factory butters presents some problems but, with the relatively uniform conditions throughout the district and the large number of cows involved in any one sample, errors from this source may be neglected. All butters were stored at 0°c. and assayed as soon as possible.

Butterfat was prepared for all assays by melting the butter, decanting the fat, and filtering at 60°c. For vitamin A and carotenoids, a sample was saponified and the unsaponifiable fraction was extracted with ether and dried. An aliquot of this was evaporated to dryness under vacuum over a water bath, the residue dissolved in chloroform, and vitamin A estimated using the Carr-Price reagent in an Evelyn-type colorimeter standardized against vitamin A acetate (L. Light & Co. Ltd.). The balance of the ether extract was also taken to dryness, and the residue was dissolved in petroleum ether (boiling range 40–60°c.) and separated into carotene and xanthophyll fractions by chromatography on alumina, essentially as described by Thompson, Ganguly and Kon[11] for the separation of vitamin A alcohol and ester. Carotene and xanthophylls were measured at 450 mμ on a Beckman spectrophotometer taking E as 2500 for 1% solution, using a 1-cm. light path.

To compare total vitamin A potencies of the fats, the carotene and vitamin A figures were converted to international units using the factors;

Carotene, I.U./g = μg/g × 1.67

Vitamin A, I.U./g = μg/g × 3.33[12]

The method for tocopherol was essentially that described by Quaife[13]. Some difficulty was experienced with the method as originally described, the main problem being a tendency for the cyclo-hexane-ethanol mixture

containing the fat to separate into two layers. This tendency was often little more than a slight turbidity in the solution, which was masked by the catalyst, but whenever it did occur hydrogenation was slow and incomplete. The problem was overcome by using absolute ethanol and limiting the concentration of fat to not more than 10% in the 1: 1 cyclo-hexane-absolute ethanol solution in the hydrogenation step. Hydrogenation was still slow and usually required 10 to 12 minutes for completion.

Iodine values were determined by the standard method[14] using the Wijs reagent.

Weather records were obtained from the Grasslands Division, Department of Scientific and Industrial Research, Palmerston North.

Throughout the season, a number of grab samples of reasonably typical Manawatu dairy pastures and a number of individual ryegrass and clover samples were taken for carotene assay. These were estimated using a modification[15] of the method of cold extraction with a "foaming mixture" of petroleum ether and ethanol as described by Moore and Ely[16].

Results

Seasonal trends found for vitamin A, carotene, xanthophylls, tocopherol, total vitamin A potency, and iodine number are shown in Fig. 1. For vitamin A, carotene, total vitamin A potency, and iodine value, these are similar to those already reported[1, 4, 5], and it is clear that the levels of tocopherol and xanthophylls in the butterfat show a similar seasonal variation.

Over the whole period, the following correlations were established between the levels of the various substances in the milk fat (25 d.f. in each case);

Carotene with vitamin A	+ 0.643	H.S.
„ „ xanthophylls	+ 0.861	H.S.
„ „ tocopherol	+ 0.900	H.S.
„ „ iodine value	+ 0.563	H.S.
Vitamin A with tocopherol	+ 0.570	H.S.
„ „ iodine value	+ 0.257	N.S.
Total vitamin A potency with tocopherol	+ 0.795	H.S.
„ „ „ „ iodine value	+ 0.459	S.

No clear correlations could be established between the levels of any of the milk-fat constituents investigated and temperature or hours of sunshine either on the day prior to sampling or over weekly intervals up to three weeks prior to sampling. Significant correlations were, however, established between the levels of carotene (+ 0.382, S) and vitamin A (+ 0.600, H.S.) in the milk fat and the rainfall during the period 14 to 7 days prior to sampling.

An indication of the secretion of vitamin A, carotenoids, tocopherol, and unsaturated acids (as measured by the iodine value of the fat) is given in Fig. 2. These secretions are calculated from the levels in the milk fat (Fig. 1) and the total factory output, which was available only as thrice-monthly averages. The assumption was made that the number

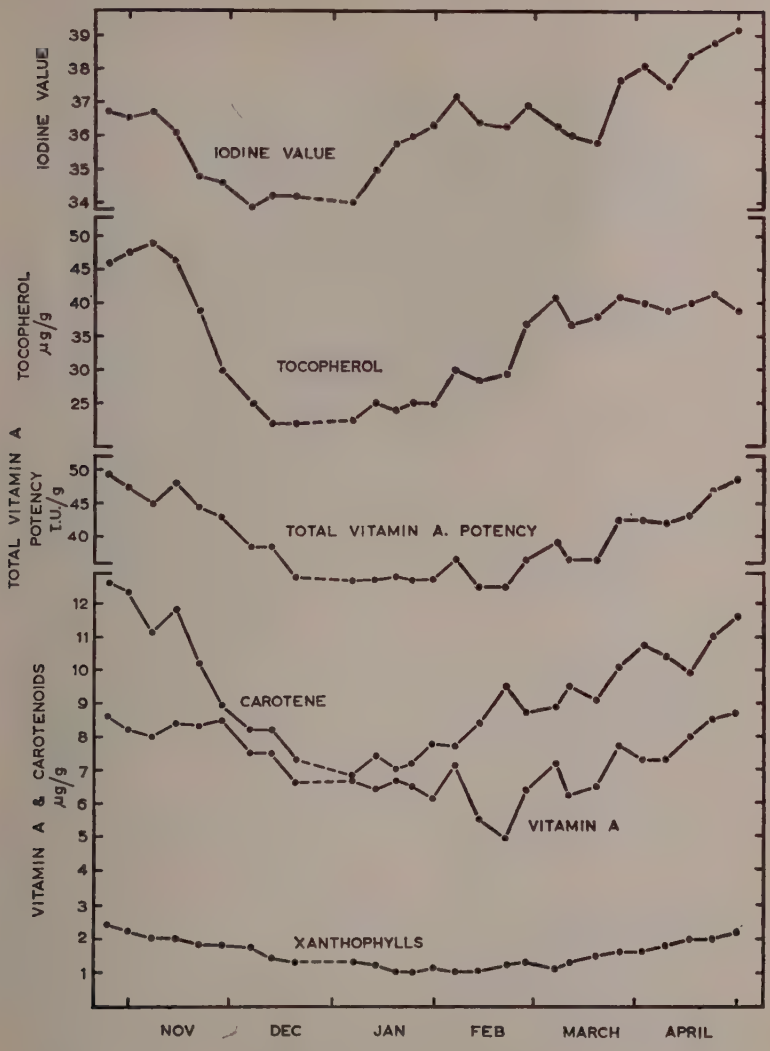

Fig. 1.—Iodine values and levels of tocophercol, vitamin A, and carotenoids in milk-fat samples.

of cows supplying the factory remained relatively constant throughout the period and, since this actual number is not known, the secretions are expressed in Fig. 2 as daily percentages of the total yield over the period covered (191 days).

Levels of carotene in the pasture showed little variation throughout the season and no seasonal trend was apparent. The average level over the whole period was 590 μg/g dry matter, and individual assays ranged from 440 to 790 μg/g. Lower levels down to 250 μg/g were found

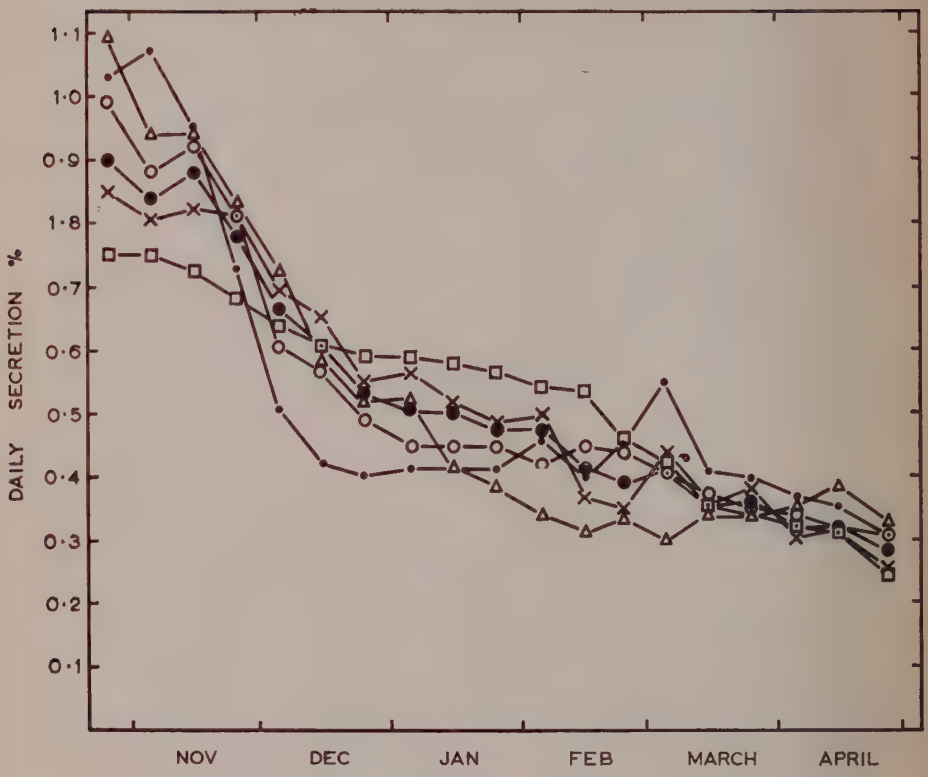

Fig. 2.—Daily secretions expressed as percentages of the total secretions over the period covered. Vitamin A X — X; carotene O — O; xanthophylls Δ — Δ; total vitamin A potency ● — ●; tocopherol · — ·; unsaturated acids (as measured by the iodine value of the fat) □ — □.

during the summer in more mature, stalky samples, but where material of this type was present in a pasture the cows were selectively grazing and the material was thus not representative of their feed. No significant differences were found between the levels of carotene in ryegrass and clover at any time during the season.

Discussion

From the results presented in Fig. 1 a clear seasonal trend is apparent in the levels of all the constituents investigated. The high level of carotene found in the pasture throughout the year confirms the previous report[9] and would give completely pasture-fed cows a daily intake of about 6 g carotene, which, even allowing for some reduction due to supplementary feeding, is still well above the levels of intake generally considered adequate for the production of milk fat of maximum vitamin A potency[10, 17]. It can only be concluded therefore that there is a seasonal trend in the availability and utilization of the carotene from the pasture. This decreased availability and utilization would

account for the decrease in milk-fat carotene and also for the decrease in milk-fat vitamin A, since the main source of this would appear to be vitamin A of immediate dietary origin[18]. Thus there must be present in pastures during late spring, summer, and early autumn, some substance that interferes with carotene uptake. It is clear, however, from the close correlation between the levels of carotene, xanthophylls, and tocopherol shown in Fig. 1 that not only carotene but other milk-fat constituents are affected. Further, since the bulk of the unsaturated acids in the milk fat is of immediate dietary origin[19], and since the bulk of the pasture fat is unsaturated[20], it would appear that over the same period there is a decreased uptake of pasture fats. Whether this results from a decreased utilization of fats and fat-soluble materials as a result of the effect of some other factors in the pasture, or from a decreased level of fat in the pasture over the summer months due to stage of maturity or botanical composition, is not known. Since fat is required as a vehicle for their absorption, any decreased uptake of fat, whatever the reason, would result in a decreased absorption of carotenoids and tocopherol, irrespective of their level in the pasture.

Failure to establish any correlations with environmental factors such as temperature confirms the findings of Stallcup and Herman[21], and it would appear that there is no effect either directly on the animal or indirectly through the pasture. The correlation in the case of rainfall is surprising but, particularly over the summer months, the effect of rain would be apparent in the pasture one to two weeks later. Stage of maturity and botanical composition of the pasture might be expected to be affected and these changes, although not associated with changes in the carotene content of the pastures nevertheless apparently influence the vitamin A potency of the milk fat produced by cows grazing them. The significance of this observation is being investigated further.

When considering variations in the levels of milk constituents, it is clear that yield must also be considered, an increased yield possibly being associated with a lower concentration and vice versa. However, the results presented in Fig. 2 show that there is a clear seasonal trend whether total daily secretion or merely concentration in the fat is considered. In the case of the daily secretions, all the constituents investigated showed similar trends, the levels falling sharply from October to late December and then more slowly until the end of lactation. In fact there is little change from February onwards, the increasing concentration of all constituents in the milk fat being offset by the decrease in fat production. This raises the question whether the increase in concentration during the autumn is a seasonal trend or an end-of-lactation effect. McDowell[5], with his work on spring and autumn calving cows, has established that the trend during spring is seasonal, but the evidence regarding the increased concentrations during the autumn is not so clear.

Acknowledgements

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CORRIGENDUM

Vol. 38 (2), August, 1956, page 152,

Introduction, line 11; *for* hypophysectomized *read* ovariectomized.

CORRIGENDA

Vol. A38(4).

Page

454. *Add footnote:* For treatments, see discussion on Factors Influencing Concentration of Soluble Salts (pp. 458, 9).

458. *Depth of Sampling* line 3, *for* 0.3 in. *read* 0-3 in.

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